Bioreactors: Design and Operation

Charles L. Cooney

Summary. The bioreactor provides a central link between the starting feedstock and the product. The reaction yield and selectivity are determined by the biocatalyst, but productivity is often determined by the process technology; as a consequence, biochemical reaction engineering becomes the interface for the biologist and engineer. Developments in bioreactor design, including whole cell immobilization, immobilized enzymes, continuous reaction, and process control, will increasingly reflect the need for cross-disciplinary interaction in the biochemical process industry.

Biotechnology can be considered to be the integration of several disciplines, including microbiology, biochemistry, molecular biology, and chemical engineering, for the purpose of utilizing biological systems for manufacturing or environmental management. Bioreactors serve a central role in biotechnological processes by providing a link between starting materials and final products, as shown in Fig. 1. It is in this area that multidisciplinary efforts are focused and value is added to a less expensive material through synthesis of a product or rendering of a service. In this article I examine the strategy for selection and design of bioreactors and identify the limits and constraints in their use. A biotechnological process is an integrated set of unit operations involving not only product synthesis and bioconversions but also product recovery. The bioreactor must be designed to meet the specific needs and constraints of a particular process, and its design will affect both cost and quality of the final product or service.

Although the term bioreactor is relatively new, the concept is quite old. Some examples of early bioreactors include the calf stomach, used in ancient times for storage of milk. It was discovered that an active ingredient in the calf stomach transformed the milk into cheese. This active ingredient is rennin, a protease that catalyzes a specific and limited hydrolysis of milk proteins and promotes their coagulation into cheese. Calf rennin is still used for cheese production. However, the bioreactor has changed from the calf stomach to temperature-controlled vats. In addition, a shortage of calf rennin has led to the use of less expensive microbially derived enzymes in some applications. Another early bioreactor was used in bread making. Bread dough provides a matrix that entraps yeast; this matrix (an early form of immobilized whole cells), placed on a flat, hot pan in an oven, traps carbon dioxide generated from the metabolic action of yeast and rises to form a loaf. With advances in technology we learned to place the dough in a pan and, while not changing the biocatalytic process, change the form and quality of the final product. The use of closed containers to promote anaerobic yeast metabolism in alcohol production is another early example of the evolution of a bioreactor to meet the needs of a particular biocatalytic process. Pasteur demonstrated the importance of oxygen for efficient production of bakers' yeast; this discovery had a considerable impact on the bakers' yeast industry and is an early example of process control. A major development in bioreactor design occurred in the 1940's, when investigators learned how to grow molds such as *Penicillium chrysogenum* in submerged culture for the production of antibiotics such as penicillin. Before this, fermentations were carried out on solid substrates, which were difficult to control and keep aseptic (*1*). Thus, the modern bioreactor in the form of a fermentor used in large-scale manufacturing of pharmaceutical products is a relatively new innovation in the fermentation industry.

The fermentation industry has expanded considerably in scope. Today it encompasses much more than fermentation and is more properly called the biochemical process industry, by analogy with the chemical process industry. A discussion of the modern biochemical process industry by Perlman (*2*) gives an overview of a multibillion-dollar industry that involves 145 companies throughout the world and produces more than 250 different products spanning the pharmaceutical, chemical, and agricultural industries. Furthermore, biological processes are used in such diverse applications as waste treatment, the production of detergent enzymes, and the enzyme-catalyzed degradation of the anticoagulant heparin in blood (*3*). Considering the central role of bioreactors in a diverse industry, it is not surprising that they are emerging in a wide variety of shapes, sizes, and forms. The goals in bioreactor design, however, are common; the purpose of a bioreactor is to minimize the cost of producing a product or a service. The evolution of bioreactors, whether for cheese production or synthesis of antibiotics, is driven by a need to increase the rate of product formation and the quality of the product or service. Bioreactor design has focused on improved biocatalysts; better process control, more recently with computers; better aseptic design and operation; and innovative ways to overcome rate-limiting steps, especially for heat and mass transfer.

Objectives in Bioreactor Design

The primary objective in the design of a bioreactor or any component of a biotechnological process is to minimize the cost of producing a high-quality product or service. Because the bioreactor provides a link between the starting materials and the product, it plays a critical role in the economics of biotechnological processes. To place this in perspective, it is useful to consider the manufacturing costs for different products. These are summarized in Table 1 for a commodity product, ethanol, and a specialty product, penicillin (*4, 5*). It is apparent that the primary cost is for the raw material and that other major costs include capital investment and utilities, mostly energy. In the case of older plants, where the capital is depreciated, maintenance changes will increase and become more significant. The magnitude of the capital investment is also a strong function of interest rates and method of financing. The cost of energy, since 1978, has risen at an annual rate of 16 percent, while raw materials and labor costs have risen at 6 and 10 percent respectively (*6*). As a

Charles L. Cooney is a professor in the Department of Chemical Engineering, Massachusetts Institute of Technology, Cambridge 02139.

result, the energy cost has become as much as 35 percent of the manufacturing cost for some products (*6*). For this reason there is increased interest in minimizing energy demands in bioreactor operation and subsequent product recovery.

Processes that require bioreactors and typical products are summarized in Table 2. It is useful to separate these processes into those that are conversion cost-intensive and those that are recovery cost-intensive; although in both cases it is important to minimize the cost of raw materials for conversion, in conversion cost-intensive processes the volumetric productivity, Q_p (grams of product per liter per hour), is of paramount importance and in recovery cost-intensive processes the product concentration, P (grams per liter), is the dominant criterion for cost minimization.

Table 1. Comparison of manufacturing costs as percent of total for penicillin (*4*) and ethanol (*5*) made by fermentation. Ethanol costs are based on corn as the starting material.

Category	Penicillin	Ethanol
Raw materials cost	35	62
Operating cost		26
Utilities	15	
Labor	4	
Maintenance	11	
Plant and overhead	8	
Capital costs	22*	12*

*Includes depreciation, taxes, and insurance.

Biological Reaction Kinetics

Several major differences between biological and chemical reaction kinetics are important in bioreactor design. In most biological systems there is a complex series of reactions that must be optimized and coordinated. Since the cell is able to facilitate this coordination in an elegant manner, growing cells are often used as a means of synthesizing complex metabolites, proteins, polysaccharides, and other products. Fermentation processes are autocatalytic, and the catalyst concentration as well as the environment changes with time; this leads to some unique process control problems. Furthermore, the reactions are conducted under moderate conditions of *p*H (near neutrality) and temperature (for instance, 20° to 65°C). While low temperature is often cited as an advantage of biocatalysis, it frequently causes a problem because heat removal is difficult at low temperature (*7*). Essentially all processing is carried out in an aqueous phase and product streams are relatively dilute (typically 0.2 to 20 percent), making recovery operations expensive.

Despite the apparent complexity of biocatalytic processes, there is usually a single rate-limiting step controlling the reaction as well as a few secondary limitations, and these rate limitations provide a basis for process design. The rate limitation may be biologically imposed, as is often the case in complex reaction sequences for antibiotic, organic acid, or amino acid synthesis, where a single enzyme step may limit metabolic flux. Productivity may be improved either by genetically alleviating the rate-limiting step in the cell or increasing the cell concentration. This is exemplified in Fig. 2, which shows a plot of Q_p versus cell or enzyme concentration, X, in a bioreactor. Productivity increases with increasing catalyst concentration and specific activity or productivity, S_a, which reflects the catalytic capacity of the cell or enzyme. Increasing the catalyst concentration is generally a biochemical engineering problem, while improving specific activity is generally a biological one. Productivity limitations may be externally imposed, either by an engineering constraint such as mass and heat transfer or by the need to limit a substrate concentration. With growing cells, limitation of a nutrient such as carbon, nitrogen, or phosphate may be required to overcome catabolite repression problems, in which one of those nutrients, when above a critical concentration, prevents synthesis of enzymes required for product formation (*8*). With an enzyme, substrate limitation may be required to avoid substrate inhibition.

The goal in bioreactor design is to minimize cost. Maximizing volumetric productivity allows one to minimize capital investment. For this reason it is usually desired to operate a reactor at its maximum physical limits, which are often constrained by mass or heat transfer (*9*). The superposition of this engineering constraint on top of biological capability is shown in Fig. 2 by dotted lines, which represent the operating limits for a bioreactor. The vertical line is the maximum catalyst concentration, which is about 200 grams per liter for cells or cellular organelles that have a moisture content of 80 percent and are packed tightly with little interstitial void volume; for enzymes it is the maximum protein loading times the specific activity. The horizontal line represents a heat or mass transfer limitation that is stoichiometrically related to product formation. When the specific activity or productivity of a microorganism or biocatalyst is improved, less catalyst is required to achieve the maximum productivity, $Q_{p\ max}$, for the reactor. This will be reflected in a reduced catalyst cost. Improvements in engineering design raise the value of $Q_{p\ max}$. Thus, even though engineering constraints limit productivity, improvements in the biological system can have a substantial impact on the cost of bioconversion.

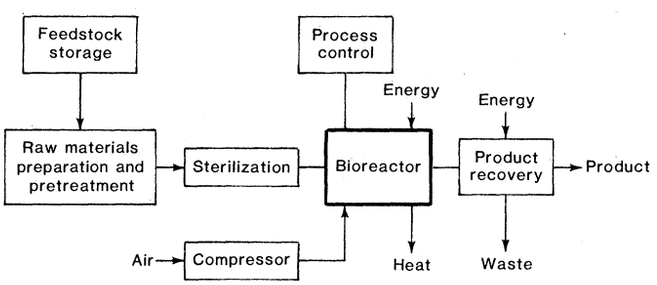

Fig. 1. Schematic overview of a biotechnological process showing the central role of a bioreactor in linking starting materials and the final product. This overview is typical of aerobic fermentation processes and unit operations such as air compression and medium sterilization may not be required in all processes.

Table 2. Examples of products in different categories in the biochemical process industry.

Category	Example
Cell mass*	Bakers' yeast, single-cell protein
Cell components†	Intracellular proteins
Biosynthetic products†	Antibiotics, vitamins, amino and organic acids
Catabolic products*	Ethanol, methane, lactic acid
Bioconversion*	High-fructose corn syrup, 6-aminopenicillanic acid
Waste treatment	Activated sludge, anaerobic digestion
Service	Heparin degradation, optical isomer resolution

*Typically conversion or feedstock cost-intensive processes. †Typically recovery cost-intensive processes.

Types of Bioreactors

In view of the importance of engineering constraints and the need to integrate biological and engineering improvements, it is useful to consider the forms of bioreactors that are commonly used today. There are three major types of bioreactors and two forms of biocatalysts. Bioreactors may be batch, semicontinuous (fed-batch), or continuous reactors; furthermore, continuous bioreactors may be continuous stirred tank reactors (CSTR) or plug flow reactors. The biocatalysts may be individual enzymes, used as soluble or immobilized catalysts, or whole cells, and the cells may be in a growing or nongrowing state. Combining these types of reactors and catalysts provides the choice of operating systems summarized in Fig. 3. The literature on novel and alternative bioreactor configurations is quite extensive (*10*); however, to tap this body of knowledge and design a bioreactor to meet specific needs requires a few simple decisions. The first is the choice between enzymes and whole cells as the biocatalyst. Today, the use of one or a few enzymes in cell-free form is limited to simple and generally degradative—that is, hydrolytic or oxidative—reactions which do not require energy coupling. Although in vitro biosynthesis, which requires energy coupling, has been demonstrated in the laboratory with cell-free enzymes (*11*), it is generally more practical to use whole cells whenever there are requirements for cofactor regeneration or energy coupling. When using enzymes, one can choose between soluble and immobilized forms that can be employed in a variety of batch and continuous reactors. The design of immobilized enzyme systems has been reviewed (*12*) and a variety of immobilization methods based on physical entrapment, covalent cross-linking, covalent attachment to a matrix, and adsorption have been described (*13*). Immobilized enzyme processes used commercially are summarized in Table 3 (*14*).

When products or services require more complex reaction sequences, it is best to use whole cells. These may be either growing cells, as used in the synthesis of most fermentation products today, or nongrowing cells, which can be used in concentrated suspensions or as immobilized cells for the catalysis of relatively simple reactions. Normally, with nongrowing cells the catalysts are immobilized by incorporation into a matrix or retention by a membrane. Such systems are being increasingly used for industrial applications, as shown by some examples in Table 3. While the biochemical process industry is characterized primarily by the use of growing cells in aqueous systems, some recent work has been done with growing immobilized cells. The apparent contradiction between growth and immobilization is resolved when the latter is considered as a form of cell recycle. Immobilization by entrapment in a gel or adsorption on a support provides a form of internal cell recycle. Alternatively, cell recycle can be achieved by devices external to the bioreactor, such as centrifugal separators, membrane devices, or settling tanks, which collect concentrated cells from a fermentation effluent and return them to the fermentor. It is more common today, however, to use growing cells in batch and semicontinuous or fed-batch fermentors, but this will not necessarily be the case in the future.

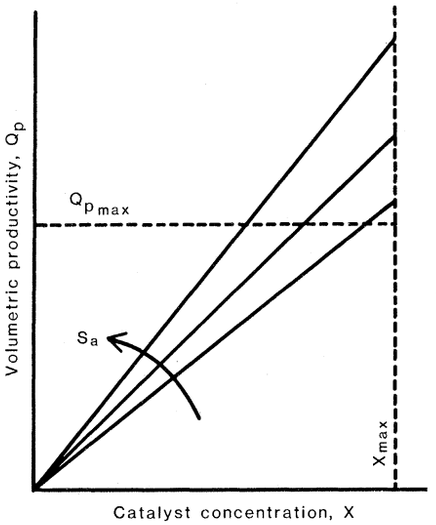

Fig. 2. Volumetric productivity (grams of product per liter per hour) in a bioreactor as a function of catalyst concentration (grams per liter) and specific activity (grams of product divided by grams of catalyst per hour). Physical constraints on productivity are shown by dashed lines and are discussed in the text.

Approaches in Bioreactor Design

A bioreactor is a biocatalyst in a container. Its engineering design is based on minimizing process constraints such as heat and mass transfer and allowing optimal control of biocatalytic activity while minimizing total process cost. Since a major cost in most biological processes is that of the raw materials, an important objective in bioreactor design is to maximize the conversion yield of starting materials to final product, $Y_{p/s}$ (grams of product divided by grams of substrate), and this reflects the catalyst selectivity. Selectivity is important not only for maximizing conversion yield but also for minimizing by-product formation, which increases problems of product recovery. While the maximum conversion yield is constrained by thermodynamics (*15*), the actual yield is constrained by bioconversion mechanisms and the requirement for biocatalyst synthesis. The objective function to be maximized is $R_{p/x}$, the ratio of product per unit of biocatalyst,

$$R_{p/x} = Y_{p/s} \int_0^{t_c} S_a(t)\, dt \qquad (1)$$

where $S_a(t)$ is the specific activity as a function of time for the cell or enzyme catalyst (grams of substrate reacted divided by grams of catalyst per unit time) and t_c is the effective catalyst life (often expressed as a half-life). Values of $R_{p/x}$ can vary greatly for different processes. For enzyme-catalyzed reactions, such as high-fructose corn syrup (HFCS) production with immobilized glucose isomerase, this ratio can be $\geq$ 2500 g per gram of enzyme preparation. For commodity products, such as single-cell protein and ethanol, $R_{p/x}$ varies from 1 to 10 g of product per gram of cells, respectively. For specialty products, such as penicillin, values of about 1 g per gram of cells are typically observed.

In the case of enzyme-catalyzed reactions, improvements in $R_{p/x}$ are made by using enzymes with high S_a, increasing the enzyme stability (t_c), and increasing the yield of active enzyme per unit mass of organism used for enzyme production. In the biosynthesis of fermentation products, $R_{p/x}$ is improved by controlling the cell's use of raw materials (primarily the carbon source) for the synthesis of cell mass, the desired product, by-products, and cell maintenance in order to maximize $Y_{p/s}$ (*9*). This can be done genetically, environmentally, or by reusing the biocatalyst as many times as possible as in cell recycle or immobilization systems. Solutions to the problem of maximizing $R_{p/x}$ involve a combination of biological and chemical engineering approaches to increasing specific activity, selectivity, and effective catalyst life.

As shown in Fig. 2, bioreactors are designed and operated to achieve a maximum volumetric productivity. The importance of productivity is reflected in the need to minimize capital investment; even if a biological process has attractive overall economics, it is difficult to achieve a high return on investment if the capital investment per unit weight of product is exceptionally high. Volumetric productivity is improved by increasing the specific activity of the enzyme or specific productivity of the growing cell

and increasing the concentration of enzyme or cells (or both) in the reactor. The use of immobilized enzymes makes it possible to achieve high catalyst concentrations in the reactor and to reuse the catalyst many times. With soluble enzymes the catalyst capacity of the reactor is often limited by the solubility of the protein catalyst and it is difficult to recover the catalyst after use. With immobilized enzymes it is possible to exceed this solubility limit since the catalyst is coupled to a solid support matrix. Then catalyst loading becomes limited by the specific activity of the immobilized enzyme preparation and the loading capacity of the immobilizing matrix. Immobilized enzyme systems such as packed columns may have protein concentrations of 50 g/liter (*16*). The specific activity of enzymes can vary (*17*) from 1 to 10,000 units per milligram of protein (where 1 unit is 1 micromole of substrate reacted per minute under optimal conditions). With highly active enzyme preparations, one could expect to use biocatalysis to achieve product formation rates greater than 100 moles per liter per hour. This is difficult to achieve, however, because of engineering limitations and physical constraints.

The volumetric productivity in fermentation or immobilized whole cell systems is limited by cell density (recall that the moisture content of cells is typically 80 percent) and by the concentration of a rate-limiting enzyme in a metabolic pathway. Bioreactors containing immobilized *Escherichia coli* and the enzyme aspartase can synthesize 3 moles of aspartic acid per liter per hour in 98 percent yield from 1.0*M* ammonium fumarate (*18*). Immobilized cell reactors containing *Saccharomyces cerevisiae* or *Zymomonas mobilis* can produce ethanol at rates greater than 1.5 moles per liter per hour (*19*). By comparison, with fermentations for antibiotics such as penicillin

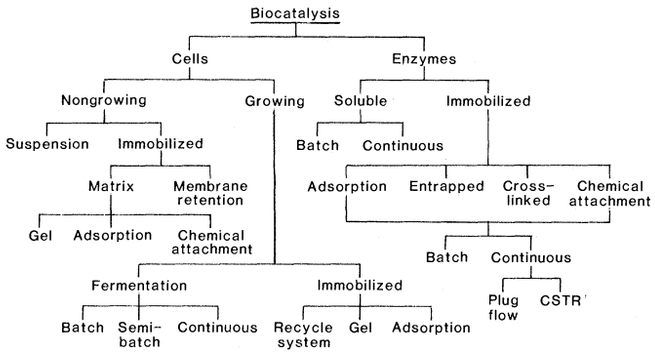

Fig. 3. Decision tree showing the options in selecting biocatalysts and bioreactor configurations.

the volumetric productivity is less than 1 millimole per liter per hour (*20*), and the chemical industry often achieves 1 to 10 moles of product per liter per hour.

Once a product is formed, it must be isolated and purified. The cost of recovery is, in general, proportional to the amount of water that must be processed. Thus it is important to maximize the concentration of product leaving the bioreactor in order to minimize both the capital investment and the operating cost of the recovery system. Antibiotics, enzymes, and, more recently, proteins produced by genetically engineered cells are examples of recovery cost-intensive products because they are often formed in dilute solution or coproduced with similar materials from which they must be purified. Recovery of highly purified intracellular protein from bacteria for therapeutic use is an example (*21*).

In a continuous process, the relation between product concentration, P, and operating conditions is

$$P = \frac{S_a V X}{F} \qquad (2)$$

where V is the volume of the reactor and F is the flow rate through the reactor. Product concentration can be maximized by decreasing the flow rate or increasing the activity and concentration of the catalyst. In the traditional fermentation industry—exemplified by antibiotic, amino acid, and organic acid production—batch and fed-batch processes are commonly used, in part because they allow high product concentrations to accumulate by letting the flow rate approach zero. Continuous processes often have higher volumetric productivities than batch processes, but the product concentration is lower. This limitation can be overcome by using high concentrations of active catalyst; examples are the use of immobilized enzymes in the conversion of penicillin to 6-aminopenicillanic acid, the conversion of glucose to HFCS, and the use of cell recycle in the conversion of sugar to ethanol.

There are other criteria to be considered in bioreactor design. Product quality must be maximized, and this is often achieved by increasing process reproducibility and minimizing by-product formation. Energy costs for air compression, mixing, temperature control, and steam sterilization have become increasingly important. The major energy cost associated with a bioreactor is that for aeration and mixing. As a consequence, considerable effort over the past 30 years has gone into the design of mass transfer systems for bioreactors that can achieve high mass transfer rates at good energy efficiency. In most biochemical processes the labor cost is relatively low, less

Table 3. Commercial immobilized enzyme reactors (*14*).

Enzyme	Product	Immobilizing method	Reactor type	Operating mode	Company	Starting date
Aminoacylase	L-Amino acids	Adsorbed	Packed bed	Continuous	Tanabe Seiyaku	1969
Aspartase*	Aspartate	Entrapped	Packed bed	Continuous	Tanabe Seiyaku	1973
Fumarase*	Fumarate	Entrapped	Packed bed	Continuous	Tanabe Seiyaku	1974
Glucose isomerase	HFCS†	Adsorbed	Packed bed	Continuous	Clinton Corn	1972
		Covalent	Stirred tank	Batch	Novo	1974
			Packed bed	Continuous	Novo	1975
Lactase	Lactase-free milk	Entrapped	Stirred tank	Batch	Snamprogetti	1977
Penicillin acylase	6-Amino penicillanic acid	Adsorbed	Stirred tank	Batch	Squibb	1966
		Covalent	Stirred tank	Batch	Astra	1973
		Covalent	Stirred tank	Batch	Beecham	1974
		Entrapped	Packed bed	Continuous	Snamprogetti	1975
Steroid dehydrogenase*		Heat-treated			Squibb	1964

*Immobilized cells. †High-fructose corn syrup.

than 5 percent of the manufacturing cost. This is achieved by designing equipment that is simple to operate, has minimal maintenance requirements, and has on-line process control. It is a characteristic of many biological processes that the equipment, particularly the bioreactor, is versatile and can be used for many different products. This is generally not true in the chemical industry, but is important in biochemical process development since often a single plant may be used for several different products.

Limitations in Bioreactor Operation and Design

While bioreactors can be loaded with sufficient catalyst to achieve very high rates of product formation, there are physical constraints in the design and operation of the reactors that prevent one from realizing these rates. Catalyst concentration and specific activity can be improved by using high cell or enzyme concentrations and catalysts with high specific activity. Ultimately, however, the rate is limited by the problem of mass or heat transfer or both. The problem of mass transfer is particularly important in processes that require oxygen but also occurs when there is a water-insoluble nutrient requirement. Equation 3 illustrates the supply and demand requirements for oxygen by a growing population of microorganisms:

$$\frac{\mu X}{Y_{O_2}} = k_L a\,(C^* - C_L) \qquad (3)$$

where μ is the specific growth rate, X the cell concentration, and Y_{O_2} the oxygen yield for growth and product formation. The rate of oxygen transfer is limited by the liquid film around the air bubble (*9*). As a consequence, oxygen transfer rates are improved by increasing the bubble surface area, a; maximizing the liquid film mass transfer coefficient, k_L; and maximizing the driving force, $C^* - C_L$, where C^* and C_L are the equilibrium concentration of oxygen in the bubble and the concentration of dissolved oxygen, respectively. Many fermentor configurations have been proposed to improve the rate and efficiency of mass transfer (*22*). The demand term is controlled by the metabolic rate of the microorganisms as characterized by the specific growth rate, μ; the density of actively growing cells, characterized by X; and the efficiency with which oxygen is coupled to energy generation and biosynthesis, as given by the value of Y_{O_2}. In most aerobic bioreactors oxygen transfer is the rate-limiting step during part or all of the process, as seen by solving Eq. 3 for X:

$$X = \frac{Y_{O_2}}{\mu} k_L a\,(C^* - C_L) \qquad (4)$$

In addition to improvements in equipment design, it is possible to use process control strategies to cope with the oxygen transfer limitation. By reducing μ during periods of high oxygen demand, it is possible to operate at a higher cell density and at the maximum productivity as determined by mass transfer rates, but to continue to accumulate and produce product in the reactor for longer periods of time. This can be done by reducing the temperature of the process or by controlling metabolism by restricting the flow of some essential nutrient such as carbon, nitrogen, or phosphate. The imposition of this restriction is important and allows C_L to be maintained above a critical value to avoid oxygen starvation of the culture, which often leads to decreased product formation and conversion yield. An important aspect of bioreactor design is integration of the design with operating strategy, and this has been a major driving force for investigations into the use of computer control in fermentation processes. In the application of process control, the fermentation industry lags many years behind the chemical process industry (*23*). This is because it is often difficult or impossible to measure the primary objective parameter—product formation—in a bioreactor and because the use of this information to control the physiology of the culture and achieve process optimization is poorly understood. In addition, biological processes are generally operated in a batch or fed-batch mode, whereas in the chemical industry continuous processes are more common. These factors have slowed the implementation of computer control in biological processes; however, process improvements through modification of microorganisms and better control are likely to provide major improvements in bioreactor operation.

Biological reactions are generally carried out at relatively low temperatures, 20° to 65°C. As a consequence, heat removal is often a problem, particularly for large bioreactors. Heat production increases linearly with volume; however, on scale-up, the surface-to-volume ratio increases by the two-thirds power. Since heat is removed in proportion to the available surface area, there is a maximum reactor size that can be cooled without mechanical refrigeration, and thus heat removal often limits the scale-up of large bioreactors. Improving the conversion efficiency for raw materials to products generally reduces the heat load (*24*); thus the problem of heat transfer limitation can be attacked by biological as well as mechanical means.

Trends and Future Directions in Bioreactor Development

It will continue to be important to develop catalysts with high specific activity and selectivity. This may be achieved at the molecular level by using genetic engineering techniques to alter the primary structure of enzyme catalysts. An alternative is the use of "artificial enzymes" as recently described by Breslow (*25*). This problem will also be attacked with better methods of enzyme immobilization, which will lead to higher retention of activity and longer lifetimes. For fermentation or whole cell systems with complex regulated and coordinated pathways, increases in specific activity can be achieved by applying recombinant DNA techniques to increase the production of rate-limiting enzymes and alter metabolic regulatory schemes in order to maximize the flow of raw materials to desired products and away from undesired by-products. Such efforts, coupled with improved process control of fermentations and improved stability of immobilized cell systems, will result in substantially lower catalyst costs.

However, bioreactor performance is usually ultimately limited by heat and mass transfer capabilities. As a consequence, there is a need to improve methods and equipment for heat and mass transfer in biological reactors. As the ability to increase catalyst loading in the reactor improves, the demands for process technology to remove process heat and transfer nutrients through diffusion barriers will increase.

The processing of large volumes of water after product removal is expensive. To minimize this cost, especially with commodity products such as ethanol and single-cell protein, water recycle is being employed. As a means of minimizing energy use, some processes are being optimized by minimizing water use, and this trend is likely to increase.

In an effort to further minimize capital investment in biotechnological processes, there will be increased emphasis on continuous processes. A major deterrent to the use of continuous bioreactors is the problem of low product concentration. This problem can be minimized by using catalysts with high specific activi-

ty. Keeping in mind the limits in carrying out a fermentation, one can envision immobilized whole cell systems providing an opportunity to achieve high productivities and hence high product concentrations. The unit operations in a biological process have a considerable effect on each other, and it is axiomatic that whatever you do upstream will have an impact on downstream processing. For this reason, it is important to carefully integrate bioreactors and all subsequent product recovery operations. Biotechnology holds great promise for the efficient production of pharmaceutical, chemical, and agricultural products. The continued success of biotechnology depends significantly on the development of bioreactors, which represent the focal point for interaction between the life scientist and the process engineer.

References and Notes

1. E. J. Lyons, *AIChE Symp. Ser.* **100**, 31 (1970).
2. D. Perlman, *Am. Soc. Microbiol. News* **43**, 83 (1978).
3. R. Langer, R. J. Linhardt, S. Hoffberg, A. K. Larsen, C. L. Cooney, D. Tapper, M. Klein, *Science* **217**, 261 (1982).
4. R. W. Swartz, *Annu. Rep. Ferment. Processes* **3**, 75 (1979).
5. Mitre Corporation, *Comparative Economic Assessment of Ethanol from Biomass* (NTIS report HCP/ET-2854, National Technical Information Service, Springfield, Va., 1978).
6. T. Murphy and S. Walsh, paper presented at the American Chemical Society meeting, Kansas City, Mo., September 1982.
7. C. L. Cooney *et al.*, *Dev. Ind. Microbiol.* **18**, 255 (1977).
8. J. F. Martin and A. L. Demain, *Microbiol. Rev.* **44**, 230 (1980).
9. D. I. C. Wang, C. L. Cooney, A. L. Demain, P. Dunnill, A. E. Humphrey, M. D. Lilly, *Fermentation and Enzyme Technology* (Wiley, New York, 1979).
10. B. Atkinson, *Biochemical Reactors* (Pion, London, 1974); S. Aiba, A. E. Humphrey, N. F. Millis, *Biochemical Engineering* (Academic Press, New York, ed. 2, 1973).
11. J. G. Stramondo *et al.*, *AIChE Symp. Rev.* **74** (No. 172), 1 (1978).
12. R. A. Messing, *Immobilized Enzymes for Industrial Reactors* (Academic Press, New York, 1975).
13. I. Chibata, *Immobilized Enzymes* (Wiley, New York, 1978); O. R. Zaborsky, *Immobilized Enzymes* (CRC Press, Boca Raton, Fla., 1973); K. Morbach, *Methods Enzymol.* **44** (1976).
14. M. D. Lilly, *DECHEMA-Monogr.* **82**, 1693 (1978).
15. V. K. Eroshin and I. G. Minkevich, *Biotechnol. Bioeng.* **24**, 2263 (1982).
16. K. Buchholz, *DECHEMA-Monogr.* **84**, 1724 (1979).
17. Th. E. Barman, *Enzyme Handbook* (Springer-Verlag, New York, 1969), vols. 1 and 2.
18. M. Fusee, W. E. Swann, G. J. Calton, *Appl. Environ. Microbiol.* **42**, 672 (1981).
19. R. R. Bland *et al.*, *Biotechnol. Lett.* **4**, 323 (1982).
20. C. L. Cooney, *Process Biochem.* **14** (No. 5), 13 (1979).
21. D. V. Goedell *et al.*, *Nature (London)* **287**, 411 (1980); T. Staehelin, D. S. Hobbs, H. Kung, C.-Y. Lai, S. Retska, *J. Biol. Chem.* **256**, 9750 (1981).
22. M. Moo-Young and H. Blanch, *Adv. Biochem. Eng.* **19**, 1 (1981).
23. C. L. Cooney, *Biotechnol. Bioeng. Symp.* **9**, 1 (1979).
24. ———, in *Microbiological Growth on C_1-Compounds*, (Society of Fermentation Technology, Japan, 1975), p. 183.
25. R. Breslow, *Science* **218**, 532 (1982).

Production of Feedstock Chemicals

T. K. Ng, R. M. Busche, C. C. McDonald, R. W. F. Hardy

Summary. Renewable raw materials may be converted by biological means to feedstocks for the chemical industry. Glucose from cornstarch is the current choice as a substrate, although advances may enable the use of less expensive lignocellulosic materials. The production of oxychemicals and their derivatives from renewable resources could amount to about 100 billion pounds annually, or about half of the U.S. production of organic chemicals. Ethanol produced by fermentation is now cost-competitive with industrial ethanol produced from fossil fuel. Biological routes to other oxychemicals exist and are expected to be important in the future. Several product recovery methods may be used, but new energy-conserving methods will be needed to make the engineering-biology combinations economical.

Photosynthetic products accumulated over eons as natural gas, petroleum, coal, and related carbonaceous materials have provided an immense resource for the development of our highly industrialized and energy-intensive civilization. However, we now recognize that this reservoir of fossil carbon is finite and is already limiting industrial growth. We must develop ways to rely more heavily on products of current photosynthesis for the basic feedstocks required by the chemical industry. Conversion of the chemical industry from a fossil carbon base to a renewable carbon base is not likely to be rapid or easy as viewed by one generation. However, it is important for future generations that we give attention and support to initiating and carrying out this conversion.

There are three major approaches to the transformation of biomass to useful feedstock chemicals: chemical, physical, and biological. Hydrogenation, pyrolysis, and fermentation are respective examples. In this article we review the current status and potential of fermentative processing of biomass to feedstock chemicals (*1–3*). We discuss the nature, supply, and economics of biomass; the microbial transformation of biomass to desired chemicals by fermentation processes; and the recovery and purification of these materials from the dilute aqueous solutions in which they are produced. We do not deal with fermentation routes to chemicals for energy use, such as ethanol. Ethanol may play a useful role as an octane enhancer in gasoline, but the amount potentially available from biomass would not be a significant replacement for fossil fuel. Fermentation processes for specialty chemicals such as antibiotics and vitamins (*4*) are well established and are not reviewed in this article.

Biomass Sources

Biomass consists of collectible plant-derived materials that are abundant, inexpensive, and potentially convertible to feedstock chemicals by fermentation processes. Biomass is found as starch in corn, wheat, potatoes, cassava, the sago palm, and other agricultural products and as monomeric sugars or soluble oligomers in corn syrup, molasses, raw sugar juice, sulfite waste liquors, and so on. It also occurs as lignocellulose in the

T. K. Ng is a research biologist, R. M. Busche planning consultant, C. C. McDonald research manager, and R. W. F. Hardy director of life sciences in the Central Research and Development Department, E. I. du Pont de Nemours & Co., Wilmington, Delaware 19898.