A Survey Course

BIOCHEMICAL ENGINEERING FUNDAMENTALS

J. E. BAILEY
University of Houston
Houston, Texas 77004
and
D. F. OLLIS
Princeton University
Princeton, New Jersey 08540

MICROBIAL AND ENZYMATIC activities have been an intimate part of man's history. Microbes probably account for greater than ninety percent of all animal mass; their biochemical action contributes significantly to chemical processes found in agriculture, diseases, digestion, antibiotic production, food manufacture and processing, spoilage, sanitation, waste disposal, and marine and soil ecology. Consequently, it is remarkable that the study of biochemical processes is not an established component of chemical engineering education.

One factor which seems to contribute to the neglect of biochemical engineering courses in many departments presently is the tendency in current texts and monographs to concentrate on a particular class of applications such as fermentations, enzyme utilization or wastewater treatment. While each such topic is important, it appears that only a course aimed at all aspects of biochemical engineering applications is likely to provide a sufficiently broad learning base to justify incorporation into most ChE curricula.

When confronted with many pages of descriptive text on biochemistry and microbiology, . . . overcoming student apprehension is perhaps the greatest challenge in teaching the course.

We have minimized this problem in our course by stressing underlying common fundamentals and a very broad range of applications. The fundamentals comprise those particular topics which profoundly influence the behavior of man-made or natural microbial or enzyme reactors. Such biological examples include the dependence of enzyme (and thus microbial) activity on substrate concentration, pH, temperature, and ionic strength, the existence of a small number of important metabolic paths among the multitude of microbial species, the cellular control mechanisms for complex internal reaction networks, and molecular devices for biological information storage and transmittal. Useful topics chosen from chemical and engineering sciences are the energetics of isothermal, coupled reactions; mixing; transfer of heat and molecular solutes; ideally and imperfectly mixed chemical reactors; and filtration.

The general character of these fundamentals is subsequently stressed by applications to class examples and a wide variety of homework problems. These latter exercises include analyses of spectrophotometry, desugaring of egg white, silver recovery from photographic film, exotoxin production, enzyme electrodes, home winemaking, chlorination disinfection, detergent biodegradation, steam reaeration, anaerobic digester heat balances, production of optically pure amino acids, and soil nitrification.

A second resistance in some of the previous efforts in biochemical engineering education arises from the assumption of significant *á priori* background in the biological sciences. Many ChE students have not studied biochemistry and microbiology, yet a working familiarity with both fields is necessary in biochemical engineering. Consequently, about thirty percent of our course is devoted to a rapid survey of those elements of microbiology and biochemistry essential to understanding biochemical reactors. It is assumed at the outset that the student is unfamiliar with both topics.

During the presentation of this material, an attempt is made to relate life sciences funda-

mentals to their process implications. For example, following discussion of molecular genetics, viruses, mutation, and genetic manipulation, we investigate recent applications of microbial genetics in developing especially productive microorganisms for several fermentation processes. Searches for explanations of the improved characteristics of the mutated microbes quickly leads to consideration of metabolic control systems and membrane transport, areas which have also been examined earlier in the course.

Obviously there is an alternative to our approach: require the students to take regular courses in the biological sciences before entering the biochemical engineering course. In what are already crowded curricula, imposition of such prerequisites greatly limits the students' opportunity to study biochemical engineering, and only those students actively engaged in research in the area are likely to elect such a sequence. On the other hand, by presenting biological fundamentals integrated with engineering analyses, design principles, and applications in a single course, the subject is easily accessible to any graduate student, and indeed to any interested upper level undergraduate. Following this course, those students concentrating in biochemical engineering can and should broaden their base in the life sciences through additional, more advanced courses taught in biochemistry, microbiology, biophysics, and other related departments.

Many ChE students have not studied biochemistry and microbiology . . . Consequently, about thirty percent of our course is devoted to a rapid survey of those elements of microbiology essential to understanding biochemical reactors.

The course is summarized in Table 1. The material progresses from atomic to macroscopic dimensions; i.e., from molecular through cellular to microbial population dynamics. Applications and the associated engineering design and analysis principles are presented as soon as the necessary background life science material has been covered. Thus, enzyme isolation and applications are considered before discussion of cell metabolism, and pure culture fermentations are examined before delving into the complexities of multiple species interactions and associated applications.

With this parallel approach, the molecular→cellular→population paradigm is maintained throughout the lectures in fundamentals and applications, description and analysis.

TEACHING THE COURSE

THE COURSE HAS BEEN taught in both a single quarter and a single semester format. In order to allow coverage of all elements of the outline during this period, an extensive set of notes has been developed so that many topics can be presented in the form of outside reading assignments. Besides their use in the courses at Houston and Princeton, all or portions of these notes have been used in biochemical engineering courses at the University of California at Berkeley, Iowa State University, the University of Maryland, and the University of Virginia. A textbook derived from these notes is now in press.

We have noted that engineering students may become a bit disoriented when confronted with many pages of descriptive text on biochemistry and microbiology, each introducing one or more new terms in an expanding cascade of new vocabulary. Overcoming this student apprehension is perhaps the greatest challenge in teaching the course, and we have used a variety of strategies in concert to ameliorate the problem.

First, divertissements from the onslaught of new biological concepts and terminology are provided in both local and longer scales in the lectures and notes. The best example of the latter case is the location of topics III and IV in the course outline. In addition to the reasons given earlier, the presence of these topics in the midst of biological basics provides the engineering student a respite in the relatively familiar territory of kinetics, transport-reaction interaction, and commercial processes. Moreover, these topics show the student that the biochemistry from topic II is indeed necessary and useful, and thus motivation is instilled for digging into energetics, metabolism, and genetics in topics V and VI.

Also, practical implications of basic concepts are briefly indicated during the fundamentals discussions. One example of this approach has already been mentioned; another is the discussion of the influence of iron ion on the citric acid fermentation in conjunction with the presentation of the tricarboxylic acid cycle. At this point previous consideration of enzyme cofactor effects can also be recalled and put into a commercial process

Jay Bailey, Professor of Chemical Engineering at the University of Houston, concentrates his teaching and research activities in biochemical engineering, chemical reactor analysis, and process dynamics. A Ph.D. graduate of Rice University, Dr. Bailey received a Camille and Henry Dreyfus Teacher-Scholar Grant in 1974. He plays guitar and 5-string banjo, now struggles with piano, and also enjoys photography.

David F. Ollis, Associate Professor of Chemical Engineering at the Princeton University, received his B.Sc. at Caltech., M.E. at Northwestern and Ph.D ('69) at Stanford. He was a process research engineer at Texaco. He was a post-doctoral fellow at CNRS, Nancy, France in 1969 and on sabbatical at CNRS in Lyons, France in 1975. His research interest include heterogenious and homogeneous catalysis and biochemical engineering.

perspective. Worked examples interspersed in the course serve a similar function of breaking up the new biological material. Finally, homework exercises on all topics have been prepared, and these are regularly assigned (a collection of problems is available from the authors upon request). Combined with frequent quizzes, these provide the engineering student with regular opportunities to attempt quantitative analyses. Besides their obvious important pedagogical role, these exercises also alleviate the "culture shock" (!) of facing many new biological terms and concepts. □

TABLE 1: COURSE OUTLINE

Biochemical Engineering Fundamentals

I. A LITTLE MICROBIOLOGY
- A. Biophysics and the Cell Doctrine
- B. The Structure of Cells (procaryotic cells; eucaryotic cells, cell fractionation) (Example: Analysis of particle motion in a centrifuge)
- C. Important Classes of Microbes (bacteria; yeasts; molds, algae and protozoa)

II. CHEMICALS OF LIFE
- A. Lipids (fatty acids; fat-soluble vitamins; steroids) Example: Modification of biomembrane permeability
- B. Sugars and Polysaccharides
- C. From Nucleotides to RNA and DNA (coenzymes; RNA, DNA)
- D. Amino Acids into Proteins (polypeptides; protein structure; biological regulation)
- E. The Hierarchy of Cellular Organization

III. THE KINETICS OF ENZYME-CATALYZED REACTIONS
- A. The Enzyme-Substrate Complex and Enzyme Action
- B. Simple Enzyme Kinetics with One and Two Substrates (Michaelis-Menten kinetics; two-substrate reactions and cofactor activation)
- C. Determination of Elementary Step Rate Constants (pre-steady-state, relaxation kinetics)
- D. Other Patterns of Substrate Concentration Dependence (activation; inhibition; multiple substrates)
- E. Modulation and Regulation of Enzymic Activity
- F. Other Influences on Enzyme Activity (pH, temperature, mechanical forces)
- G. Enzyme Reactions in Heterogeneous Systems (insoluble substrates; immobilized enzymes)

IV. ISOLATION AND UTILIZATION OF ENZYMES
- A. Production of Crude Enzyme Extracts
- B. Enzyme Purification (chromatography; dialysis; solid phase syntheses)
- C. Enzyme Immobilization
- D. Application of Hydrolytic Enzymes (esterases, carbohydrases, proteases)
- E. Other Enzyme Applications (medical, new technology)
- F. Immobilized Enzyme Technology (industrial processes; medical and analytical applications; utilization and regeneration of cofactors)
- G. The Scale of Enzyme Technology

V. METABOLIC PATHWAYS AND ENERGETICS OF THE CELL
- A. The Concept of Energy Coupling: ATP and NAD
- B. Anaerobic Metabolism: Fermentation (glycolysis; other pathways)
- C. Respiration and Aerobic Metabolism (TCA cycle; respiratory chain; partial oxidation; regulation)
- D. Photosynthesis: Tapping the Ultimate Source (Calvin Cycle; chloroplasts)
- E. Biosynthesis (ATP utilization; small molecules; macromolecules)
- F. Transport Across Cell Membranes (passive, facilitated, active transport) (Example: Transport of nitric acid through a liquid membrane)

VI. CELLULAR GENETICS AND CONTROL SYSTEMS
- A. Molecular Genetics (DNA translation; replication; mutation; induction; repression)
- B. Growth and Reproduction of a Single Cell (synchonous culture; E. coli cell cycle; eucaryotic cell cycle)
- C. Alteration of Cellular DNA (viruses, phage; transformation, conjugation; composite DNA)
- D. Commercial Applications of Microbial

Genetics and Mutant Populations (Implications for medium formulation; auxotrophic mutants)

VII. KINETICS OF SUBSTRATE UTILIZATION, PRODUCT YIELD, AND BIOMASS PRODUCTION IN CELL CULTURES
- A. Growth Cycle Phases for Batch Cultivation (lag phase; exponential growth, the Monod equation; stationary and death phase)
- B. Mathematical Modeling of Batch Growth (reaction networks; structured, unstructured models; mold growth)
- C. Product Synthesis Kinetics (fermentation classifications; Shs segregated model)
- D. Overall Kinetics in Cases of Reaction-Mass Transport Interation (lumped, distributed models for cells, floes, mold pellets)
- E. Thermal Death Kinetics of Cells and Spores

VIII. TRANSPORT PHENOMENA IN MICROBIAL SYSTEMS
- A. Gas-Liquid Mass Transfer in Microbial Systems (basic concepts; metabolic oxygen utilization rates) (Example: Effectiveness factor of a microbial monolayer)
- B. Determination of Oxygen Transfer Rates (gas-liquid reactions; dissolved oxygen measurements) (Examples: Warburg respirometer; electrochemical determination of $k_l a$)
- C. Mass Transfer for Freely Rising or Falling Bodies
- D. Mass Transfer Across Free Surfaces
- E. Forced Convective Mass Transfer (key dimensionless groups; mass transfer coefficient correlations)
- F. Surface Area Correlations for Mechanically Agitated Vessels
- G. Other Factors Affective $k_l a$ (diffusivities; ionic strength; surface active agents)
- H. Non-Newtonian Fluids (models; suspensions; power consumption mass transfer)
- I. Scaling of Mass Transfer Equipment
- J. Particulate Mass Transfer: Filtration (single fiber efficiencies; mass transfer coefficients)
- K. Heat Transfer (microbial heat generation; heat transfer correlations)

IX. BIOLOGICAL REACTOR DESIGN AND ANALYSIS
- A. The Ideal Continuous Flow Stirred Tank Reactor (Monod's chemostat; incomplete mixing, films, recycle effects; enzyme catalyzed reactions) (Example: Agitated CSTR design for a liquid hydrocarbon fermentation)
- B. Residence Time Distributions (measurements; applications)
- C. Tubular and Tower Reactors (ideal plug flow tubular reactor; tower reactors; tanks-in-series and dispersion models)
- D. Sterilization Reactors (bath; continuous)
- E. Relationships Between Batch and Continuous Biological Reactors (Example: Reactor modeling and optimization for production of α-Galactosidase by a Monascus sp. mold)

X. BIOLOGICAL REACTORS, SUBSTRATES, AND PRODUCTS I: SINGLE SPECIES APPLICATIONS
- A. Fermentation Technology (medium formulation; aseptic practice; cell harvesting, product recovery)
- B. Product Manufacture by Fermentation (brewing and wine making; oxidative transformations; organic, amino acids; complex molecules: gibberelins, vitamins, antibiotics; undesirable products) (Example: Reaction rates in microbial films; temperature programming for optimal pencillin production)
- C. Reactors for Biomass Production (food; food processing; agricultural applications; immunology, tissue culture, and "Vaccine" production) (Examples: A batch growth model for liquid hydrocarbon fermentations; production of a low-intermediate molecular weight product; cell growth and virus propagation kinetics in tissue culture)

XI. ANALYSIS OF MULTIPLE, INTERACTING MICROBIAL POPULATIONS
- A. Neutralism, Mutalism, Commensalism, and Ammensalism
- B. Mathematical Preliminaries (Example: Two-Species dynamics near a steady state)
- C. Competition: Survival of the Fittest
- D. Predation and Parasitism (Lotka-Volterra model; other one predator-one prey models) (Example: Model discrimination and development via stability analysis)
- E. Effects of the Number of Species and their Web of Interactions (trophic levels, food chains, food webs; mass action models; qualitative stability; randomly constructed food webs) (Examples: An application of the mass action theory; qualitative stability of a simple food web)
- F. Spatial Patterns

XII. BIOLOGICAL REACTORS, SUBSTRATES, AND PRODUCTS II: MIXED MICROBIAL POPULATIONS IN APPLICATIONS AND NATURAL SYSTEMS
- A. Uses of Well-Defined Mixed Populations (Example: Enhanced growth of methane-utilizing Pseudomonas sp. due to mutalistic interactions in a chemostat
- B. Spoilage and Product Manufacture by Spontaneous Mixed Cultures
- C. Microbial Participation in the Natural Cycles of Matter and Energy
- D. Biological Wastewater Treatment (wastewater characteristics; activated sludge process; trickling biological filters; anaerobic digestion) (Example: Simulation studies of control strategies for anaerobic digesters)