

ADVANCES IN
BIOPROCESS
ENGINEERING

ADVANCES IN BIOPROCESS ENGINEERING

Edited by

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and

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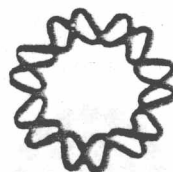
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**This book consists of papers presented at
the First International Symposium on Bioprocess Engineering
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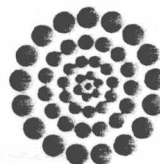
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PREFACE

The exigent and rapidly growing markets of biotechnological products are demanding new and highly efficient bioprocesses. Molecular biology and immunology techniques have been crucial for achieving the important progress that the so called new biotechnology has made. However, improvements in actual process and industrialization of new ones depend more on how well a laboratory scheme is translated to production scale. In this activity, bioprocess engineering plays a key role.

Motivated by the belief that increased participation of engineering is needed in nowadays biotechnology, biochemical engineers at the Institute of Biotechnology of the National University of Mexico in Cuernavaca, Morelos, Mexico, organized the *First International Symposium on Bioprocess Engineering*. The Symposium aimed to be an International forum to present and discuss Bioprocess Engineering issues.

The Symposium consisted of invited and referred papers. Thirteen distinguished biochemical engineers were invited to give lectures at the Symposium. In addition, the response from the international community was very enthusiastic. About 120 abstracts were submitted for consideration. From the former, 80 were selected to submit full manuscripts, which then were sent to referees for reviewing. Finally, 55 papers were accepted for presentation in the Symposium and most of them are published in this book. The Symposium was truly international, 33 countries were represented from all over the world.

The papers address several topics of bioprocess engineering, namely: fundamentals of biochemical engineering, process development and optimization, control and automation of bioprocesses, design and scale up of bioreactors, enzyme engineering, waste treatment and downstream operations.

Basic bioengineering aspects such as transport mechanisms, models for mycelial growth, oxygen transport and structured models for bioreactors, rheology and mixing in fermentations and the utility of image analysis were addressed in some of the papers. Bioprocesses for the production of ethanol, gibberellic acid, lactic acid, proteases, β -galactosidase, anakinra, streptokinase, penicillin-amidase, and lipases, as well as a bioleaching process, were described in various papers. Traditional and new processes (recombinant fermentations, plant and animal cell culture, and transgenic animals) are represented. Solid, liquid and gaseous substrates were considered in a variety of bioprocesses. The bioreactor modes studied included stirred tanks, air-lifts, percolation columns and packed and fluidized beds with immobilized cells. Interesting control problems are discussed for various model systems such as fermentors, enzymatic reactors and downstream operation systems. Authors from industry have also shared some of their experiences in process development and scale-up techniques. A good number of papers dealt with the treatment of solid, liquid or gaseous wastes using aerobic and anaerobic processes. Enzymatic engineering was well represented with examples of immobilized enzyme reactors, peptide synthesis and non-aqueous biocatalysis. Downstream operations papers dealt with the recovery of therapeutic bioproducts produced by recombinant microorganisms and transgenic animals.

The Symposium and the book would have been impossible to make without the participation of many people in various stages and aspects. We want to thank the invited speakers, the authors, the Scientific Committee and the referees for their enthusiastic response, their time and work in making this Symposium a valuable one. Our gratitude to our sponsors, who made possible the Symposium and the book. We thank the National University of

Mexico and particularly the Institute of Biotechnology for their support and encouragement in organizing this event. Many thanks to all the Organizing Committee for helping us in taking care of endless organization details and to our wives for their patience and support.

We hope this book can contribute to the development of Bioprocess Engineering, a key profession in turning biotechnology possibilities into biotechnology realities.

Enrique Galindo
Octavio R. Ramírez

Symposium Organizers and Editors
Institute of Biotechnology, National University of Mexico,
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TABLE OF CONTENTS

Preface	xv
Design, Operation and Scale-up of Bioreactors	
Bioreactor Engineering <i>N.W.F. Kossen, Gist Brocades B.V. (Invited), The Netherlands</i>	1
Design and Scale-Up of External-Loop Airlift Bioreactors <i>Y. Kawase, Tokyo Univ., Japan</i>	13
Engineering a Successful Scale-Up of a Process to Produce a rDNA Product <i>G.F. Slaff, Synergen, U.S.A.</i>	21
Slurry Bioreactor Design for Shear-Sensitive Mycoprotein Production <i>Y. Chisti and M. Moo-Young*, Univ. of Waterloo (Invited), Canada</i>	25
Strategies in the Design of a Penicillin Acylase Process <i>A. Gómez, M. Rodríguez, S. Ospina, E. Merino, F. Bolívar, O.T. Ramírez, R. Quintero and A. López-Mungía*, National Univ. of Mexico (Invited), Mexico</i>	29
Scaling-Up of a Lipase Fermentation Process: A Practical Approach <i>S.G.M. Geraats, Gist-Brocades B.V., The Netherlands</i>	41
Optimum Design of a Continuous Fermentation Unit of an Industrial Plant for Alcohol Production <i>S.R. Andrietta and F. Mangeri*, Univ. Campinas, Brazil</i>	47
Scale-Up <i>N.W.F. Kossen, Gist Brocades B.V. (Invited), The Netherlands</i>	53
High Cell Density Yeast Production: Process Synthesis and Scale-Up <i>M. de la Torre*, L.B. Flores and E. Chong, National Polytechnic Institute (Invited), Mexico</i>	67
Continuous Flow Cell-Recycle Fermentation of Biomass Hydrolysates <i>¹C.H. Choi and ²A.P. Mathews*, ¹Kwandong Univ., Korea, ²Kansas State Univ., U.S.A.</i>	75
Physiological and Technico-Engineering Aspects of Lignocellulose Solid-State Fermentation with Filamentous Fungi <i>U.E. Viesturs* and M.P. Leite, Latvian State Institute of Wood Chemistry, Latvia</i>	81
Growth of <i>Candida Utilis</i> on Amberlite with Glucose and Ethanol as Sole Carbon Sources <i>¹P. Christen*, ¹R. Auria, ²R. Marcos, ²E. Villegas and ²S. Revah, ¹ORSTOM, France, ²Univ. of Aut. Metropolitana-Iztapalapa, Mexico</i>	87

- Scaling-Up Aerobic Fermentation which Produce Non-Newtonian, Viscoelastic Broths
D.W. Hubbard*, S.E. Ledger and J.A. Hoffman, Michigan Technological Univ. (Invited), U.S.A. 95

Animal and Plant Cell Culture

- Plant Cells as Chemical Factories: Control and Recovery of Valuable Products
A.E. Humphrey, Pennsylvania State Univ. (Invited), U.S.A. 103
- Optimization of Fed-Batch Mammalian Cell Culture Processes
W. Zhou and W.-S. Hu*, Univ. of Minnesota (Invited), U.S.A. 109
- A Fluidized Bed Reactor for the Cultivation of Animal Cells
J. Keller and I.J. Dunn*, ETH-Zurich (Invited), Switzerland 115
- Influence of Time and Multiplicity of Infection on the Batch Production of *Anticarsia Gemmatalis* Nuclear Polyhedrosis Virus in Lepidopteran Insect Cell Cultures
G. Visnovsky and J. Claus*, Univ. Nacional del Litoral, Argentina 123
- Design of Tubular Microporous Membrane Aerated Bioreactors for Plant Cell Suspension Culture
A.E. Humphrey, Pennsylvania State Univ. (Invited), U.S.A. 129
- Optimization and Scale-Up of High Density Cell Culture Bioreactors
S.S. Ozturk, Miles Biotechnology, U.S.A. 133

Basic Aspects in Biochemical Engineering

- Recent Studies on Stirred Bioreactors at the SERC Centre for Biochemical Engineering at Birmingham
A.W. Nienow, Univ. of Birmingham (Invited), U.K. 141
- Role of Turbulence in Fermentations
P.K. Namdev*, E.H. Dunlop, K. Wenger and P. Villeneuve, Colorado State Univ., U.S.A. 149
- Power Input and Oxygen Transfer in Fed-Batch Penicillin Production Process
A.C. Badino, M. Barboza and C.O. Hokka*, Univ. Federal de Sao Carlos, Brazil 157
- The Examination of Bioreactor Heterogeneity with Rheological Different Fermentation Broths
D.J. Pollard*, A.P. Ison, P. Ayazi Shamlou and M.D. Lilly, Univ. College London, U.K. 163
- The Influence of Cell Concentration and Morphology on the Yielding Behavior of Filamentous Fermentation Broths
*M. Mohseni, *H. Kautola and *D.G. Allen*, *Univ. of Toronto, Canada, *Seinajoki Inst. of Tech. (Invited), Finland 171
- A New Approach for Modelling the Kinetics of Mycelial Cultures
G. Viniegra-González*, P. Larralde-Corona and F. López-Isunza, Univ. Aut. Metropolitana-Iztapalapa, Mexico 183
- On-Line Estimation of Yeast Growth Rate Using Morphological Data from Image Analysis
K. Zalewski*, P. Götz and R. Buchholz, Technische Univ. Berlin, Germany 191

Growth and Protein Formation of Recombinant <i>Aspergillus</i> : Utility of Morphological Characterization by Image Analysis	
<i>M. Carlsen¹, A. Sphor¹, R. Mørkeberg, J. Nielsen and J. Villadsen</i> , Technical Univ. of Denmark, Denmark	197
Inducing Controlled Growth of <i>Penicillium Chrysogenum</i> in Pelletized Form	
<i>S.M. Ratusznei and C.A.T. Suazo¹</i> , Univ. Federal de São Carlos, Brazil	203
Structured Modelling of Bioreactors	
<i>M. Reuss¹, S. Schmalzriedt and M. Jenne</i> , Univ. Stuttgart (Invited), Germany	207
Measurement and Modelling of Oxygen Transport into Biotechnical Immobilisates for Cell Entrapment	
<i>R. Wiesmann¹, P. Götz and R. Buchholz</i> , Technische Univ. Berlin, Germany	217
Diffusion of Phosphate Ion in Xanthan Gum Solutions	
<i>G. Araiza, L.G. Torres and E. Galindo¹</i> , National Univ. of Mexico, Mexico	221
Modelling Protease Production by Immobilised <i>Serratia Marcescens</i>	
<i>C. Quirós, L.A. García¹ and M. Díaz</i> , Univ. of Oviedo, Spain	227
A Mathematical Model for a Fermentation Process Carried out in a Fluidized Bed Reactor	
<i>A.V. González-Alvarez¹, V. Alcaraz-González, and V. Zúñiga-Partida</i> , Univ. of Guadalajara, Mexico	233
Modelling the Growth of <i>Acidothermus Cellulolyticus</i>	
<i>D.W. Hubbard¹, T.B. Co, P.P.N. Murthy and R. Mandalam</i> , Michigan Technological Univ., U.S.A.	241
Measurement, Control, and Automation of Bioprocesses	
Bioreactor coupled on-line measurements	
<i>M. Reuss¹, A. Riek, M. Schütz and W. Mailinger</i> , Univ. Stuttgart (Invited), Germany	247
An Experimental Guide to the Relevant Aeration Rates in Microaerobic Bioprocesses	
<i>C.J. Franzén, G. Lidén and C. Niklasson¹</i> , Chalmers Univ. of Technology, Sweden	255
Model Aided Multiple Correlation Analysis between Preculture and Main Fed-Batch Culture	
<i>R. Guthke¹ and W. Rausch</i> , Hans-Knöll-Institute for Natural Product Research, Germany	267
Identification Techniques for a Recombinant Fed-Batch Fermentation for Ethanol Production	
<i>V.M. Saucedo, B. Eikens and M.N. Karim¹</i> , Colorado State Univ., U.S.A.	275
Computer Controlled Enzymatic Reactor	
<i>L.A. Minim and R. Maciel Filho¹</i> , Univ. Estadual de Campinas, Brazil	283
Pressure Drop as a Method to Evaluate Mold Growth in Solid State Fermentors	
<i>¹R. Auria and ²S. Revah¹</i> , ¹ ORSTOM, France, ² Univ. Aut. Metropolitana-Iztapalapa (Invited), Mexico	289
Computer-Aided Design of Integrated Biochemical Process	
<i>¹D.P. Petrides¹ and ²J. Calandranis</i> , ¹ New Jersey Institute of Technology, U.S.A., ² Intelligen Inc., U.S.A.	295

A Strategy for the pH Control in Acidic Wastewaters <i>O. Galán-Domínguez, J. Álvarez-Ramírez* and J. Álvarez-Calderón, Univ. Aut. Metropolitana-Iztapalapa, Mexico</i>	305
Modelling and Control Strategies for the Transformation of D-Sorbitol to L-Sorbose on a Laboratory Bioreactor <i>A.T. Fleury*, A. Bonomi, E.F.P. Augusto, L.H.C. Quiroz, M.F. Barral and P.S. Pereiralima, Instituto de Pesquisas Tecnológicas do Estado de São Paulo, Brazil</i>	313
The Control Problem for the Protein Separation by the Continuous Affinity Recycling Extraction Process <i>M.I. Rodríguez, R. Maciel Filho and F. Maugeri*, Univ. Campinas, Brazil</i>	321
Bioremediation and Waste Treatment	
Biodegradation of PCP in Contaminated Soil and in the Aqueous Phase <i>J.F. González* and W.-S. Hu, Univ. Nac. de Mar del Plata, Argentina, Univ. of Minnesota, U.S.A.</i>	329
Biodenitrification Studies with a Bioreactor Operating in a Periodic Mode <i>J.-H. Wang, B.C. Baltzis* and G.A. Lewandowski, New Jersey Institute of Technology, U.S.A.</i>	337
Dynamic Behavior of Activated-Sludge in Exponentially Fed-Batch Cultures Subjected to Step Perturbations <i>O.T. Ramírez*, A. Aguilar-Aguila and R. Quintero, National Univ. of Mexico, Mexico</i>	345
Expansion Characteristics of Tapered Fluidized-Bed Bioreactors <i>C.-S. Wu and J.-S. Huang*, National Cheng Kung Univ., R. of China</i>	355
High-Rate Biofilm Fluidized Bed Reactors for Specialized Wastewater Treatment <i>I.J. Dunn, ETH-Zurich (Invited), Switzerland</i>	365
The Effect of Concentration and Hydraulic Shock Loads on the Performance of a Two-Stage High-Rate Anaerobic Wastewater Treatment System: Prediction and Validation <i>M. Romli*, J. Keller, P.L. Lee and P.F. Greenfield, Bogor Agricultural Univ., Indonesia, The Univ. of Queensland, Australia</i>	379
Investigation of Kinetic and Microbiological Features of UASB-Reactor Performance under Various Organic Loading Rates <i>S. Kalyuzhnyi*, V. Sklyar and J. Rodríguez, Autonomous Univ. of Coahuila, Mexico</i>	385
Reduction of Antiphenological Compounds of Coffee Pulp Through Solid State Fungal Fermentation <i>C. Porres*, J. Calzada and D. Alvarez, Central American Res. Inst. for Industry (ICAITI), Guatemala</i>	391
Biological Removal of Hydrophobic Solvent Vapors from Airstreams <i>Z. Shareefdeen and B.C. Baltzis*, New Jersey Institute of Technology, U.S.A.</i>	397
Toluene Removal from Air Stream by Biofiltration <i>M. Morales, F. Pérez, R. Auria and S. Revah*, Univ. Aut. Metropolitana-Iztapalapa, Mexico, ORSTOM, France</i>	405

Metabolite Production, Physiology and Microbiology

- Metabolic Engineering of Cephalosporin Biosynthesis in *Streptomyces Clavuligerus*
L.-H. Malmberg, A. Khetan, D.H. Sherman and W.-S. Hu*, Univ. of Minnesota (Invited), U.S.A. 413
- High Production of Lactic Acid from Metabolically Engineered *Saccharomyces Cerevisiae*
L. Brambilla, D. Porro*, E. Martegani, B.M. Ranzi and L. Alberghina, Univ. of Milan, Italy 417
- Some Aspects of *Gibberella Fujikuroi* Culture Concerning Gibberellic Acid Production
¹P.C. González*, ¹G. Delgado, ¹M. Antigua, ¹J. Rodríguez, ²P. Larralde, ²G. Viniegra, ³L. Pozo and
³M.C. Pérez, ¹Cuban Inst. for Res. on Sugar Cane By-prods., Cuba, ²Univ. Aut. Metropolitana-Iztapalapa, Mexico, ³Inst. de Cítricos y Frutales, Mexico 425
- Continuous Culture to Produce Recombinant β -Galactosidase in *Bacillus Subtilis*
C.A. Rincón, R. Quintero and M. Salvador*, National Univ. of Mexico, Mexico 431
- Rapid Ethanol Production from Sucrose Using *Zymomonas Mobilis*
¹L.A. Kirk and ²W.H. Doelle*, ¹Univ. of Queensland, Australia, ²Microbiotech Pty. Ltd., Australia 437
- Improved Production of Proteases by *Brevibacterium Linens* in Submerged Culture
¹S. Strauss*, ²J. Zemanovic and ¹W. Hampel, ¹Univ. of Technology Vienna, Austria, ²Slovak Techn. Univ., Slovakia 441
- Production of KI (Streptokinase) Using Batch and Fed-Batch Culture of Recombinant *E.Coli*
¹R.E. Narciani*, ²F.J. Morbe, ²U. Knupfer, ²R. Wenderoth and ²D. Riesenberger, ¹Center for Genetic Engineering and Biotechnology, Cuba, ²Hans-Knöl-Institut für Naturstoff-Forschung, Germany 447
- Microbial Protein Production by Submerged Fermentation of Mixed Cellulolytic Cultures
I. Zabala*, A. Ferrer, A. Ledesma and C. Aiello, Univ. del Zulia, Venezuela 455

Enzyme Engineering

- Chitin as a Matrix for Enzyme Immobilization
A. Illanes, Univ. Católica de Valparaíso (Invited), Chile 461
- Enzyme Reactor Performance under Thermal Inactivation
A. Illanes*, C. Altamirano and O. Cartagena, Univ. Católica de Valparaíso (Invited), Chile 467
- Process Strategies in Enzymatic Racemate Resolution
J.L.L. Rakels*, A.J.J. Straathof and J.J. Heijnen, Delft Univ. of technology, The Netherlands 473
- Design of Reaction Medium for Nonaqueous Biocatalysis: Analysis on Enzyme Hydration and Catalytic Activity in Organic Solvent
S.B. Lee*, K.-J. Kim and M.G. Kim, Pohang Univ. of Science and Technology, Korea 481
- New Approaches to the Enzymatic Production of Oligopeptides: Synthesis of the "Delicious Peptide" and its Fragments
I. Gill, R. López-Fandiño, X. Jorba and E.N. Vulfson*, AFRC, BBSRC Institute of Food Research, U.K. 485

<i>Aspergillus</i> sp. 2M1 xylanases: Production, Characterization and Application in the Pulp and Paper Industry	
¹ N. Duran*, E. Curotto, ¹ E. Esposito, ² C. Aguirre and ^{1,2} R. Angelo, ¹ Univ. E. de Campinas, Brazil, ² Univ. Católica de Valparaíso, Chile	489
Optimization of Cell Harvesting and Assay Procedures for Reductive Biotransformations in Obligate Anaerobes	
E.T. Davies* and G.M. Stephens, Univ. of Manchester, U.K.	495
Downstream Processing and Bioseparations	
A Scalable Method for the Purification of Recombinant Human Protein C from the Milk of Transgenic Swine	
¹ W.N. Drohan, ² T.D. Wilkins, ² E. Latimer, ² D. Zhou, ³ W. Velandar, ¹ T.K. Lee and ¹ H. Lubon*, ¹ American Red Cross, U.S.A., ² TechLab. Inc., U.S.A., ³ Virginia Polytechnic Institute, U.S.A.	501
A Laboratory Study on the Behavior of <i>Thiobacillus Ferrooxidans</i> during Pyrite Bioleaching in Percolation Columns	
¹ M.G. Monroy*, ² M.A. Dziurla, ² B.T. Lam, ² J. Berthelin and ³ P. Marion, ¹ Univ. A. of San Luis Potosi, Mexico, ² CNRS-Univ. of Nancy, France, ³ CNRS-Ecole Nationale Supérieure de Géologie Appliquée de Nancy, France	509
Purification of G6PDH from Unclearified Yeast Cell Homogenate using Expanded Bed Adsorption (EBA) with STREAMLINE™ Red H-E7B	
Y.K. Chang*, G.E. McCreath and H.A. Chase, Univ. of Cambridge, U.K.	519
Purification of Recombinant Hepatitis B Surface Antigen (rec-HBsAg) from <i>P. Pastoris</i> : A Process Development Study	
¹ L. Pérez*, ² S. López, ¹ A. Beldarraín, ¹ D. Arenal and ¹ E. Pentón, ¹ Center for Genetic Engineering and Biotechnology, Cuba, ² Center for Biological Productions, Cuba	527
Evaluation of Affinity Chromatographic Methods in the Purification of recombinant Streptokinase	
N. Pérez*, P. Rodríguez, L. Hernández, E. Muñoz, D.R. Orta and S. Pérez, Center for Genetic Engineering and Biotechnology, Cuba	535
In Appreciation	541

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Bioreactor Engineering

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This paper is not about formal design rules for bioreactors as such. There is plenty of literature available about this subject: Bailey and Ollis [1]; Schügerl [2]; Van't Riet [3], Kossen [4]; as well as about its limitations: Zlokarnik [5]. The continuous thread through this paper is that the kind of product to be made in the bioreactor has a direct influence upon the time and the money that is available for the design of a new reactor, or the adaptation of an existing one. Time and money determine to a large extent the rules and methods that should be used. Designs with a built-in flexibility will appear to be essential. Finally the consequences for the education of those involved in bioprocess engineering (including bioreactor design) will be discussed.

THE DEVELOPMENT OF A BIOREACTOR AS A PART OF A PRODUCT/PROCESS DEVELOPMENT CHAIN (PDC)

The development of a bioreactor is an integrated part of a chain of events, the product/process development chain (PDC) (fig.1). A number of remarks can be made regarding this PDC:

First of all the environment of the micro-organisms is different in every part of the chain. The greatest difference is between the environment during screening and production (any resemblance between these environments is a lucky coincidence). Due to the flexibility of biotechnology this problem can be overcome further down stream the PDC.

Second, the over all success of the development of a process in biotechnology depends on all the steps of this chain. Contrary to ordinary chains, however, weak links of this chain can be compensated by the contribution of other links.

If e.g. the productivity of a bioreactor is below standard this problem can be solved by a number of methods:

- a. selection of a new strain
- b. genetic improvement of the strain by either classical or molecular genetics
- c. physiological improvements like changing the composition or the rate of addition of the substrate
- d. improving the bioreactor as such
- e. improving the yield of the downstream processing.

There is a definite difference between the methods mentioned in this list. The first three, a, b, and c, are "upstream" solutions (relative to the bioreactor). Furthermore they are "software" solutions (no investments in nuts and bolts are needed). Finally results in this area can often be obtained rapidly and with relatively few investments (2 or 3 man years of work and often much less). Solution d is a typical "hardware" solution, and the investments needed can be quite substantial.

Conclusions. The conclusions of this paragraph are that there is not just one solution to a problem. It even shows, "horribile dictu," that the solution can be achieved by disciplines different from your own, and that there is no "a priori"

argument why the contribution of one discipline is better than that of another.

THE RELATION BETWEEN THE DESIGN OF A BIOREACTOR AND THE PRODUCT.

There are a number of relations between the design of a bioreactor and the product:

- a. Via the sequence: product ----> micro organism ----> reaction conditions (the "environment") ----> reactor design. This is the approach of the PDC and is mainly used for the design of a new process for a new product.
- b. Via the influence of the process on the product:
 - yield
 - properties (composition, structure of enzymes etc.)
 This approach is often used when an existing process has to be optimized.
- c. Commodities vs specialties:

commodities

-cost leader
-long life cycle
(>10y)
-large market share
(economy of scale)
-optimization
-"hard/software" sol.
-patents an issue

specialties

-differentiation
-short life cycle
(<5y)
-first to the market
(economy of time)
-"quickies"
-"software" solutions
-patents a big issue

The relations a and b are well known, and are well integrated in the teaching programs of the universities. This is not the case for relation c. The focus will from now on be on the similarities and the differences between the development of bioreactors for commodities and specialties.

Commodities

The main point here is to realize a lower cost price than your competitors. This usually asks for economy of scale (for production and marketing). We will restrict our attention to the influence on the design of bioreactors.

A low cost price usually means (among others) a very sophisticated design

of a reactor (low fixed and variable costs). This often results in a design where the process fits in the installation as a hand in a glove. If this fit is not proper, costly adaptations are necessary (as an example the adaptations in the ICI bioreactor for the production of "pruteen" to increase the mixing performance can be mentioned). Furthermore the collection of all the necessary data (especially those on kinetics) can be extremely elaborate. For the production of commodities in biotechnology this close fit is not necessarily the right philosophy as will be shown in the next paragraph.

Quite often the successes of the "software" solutions (improvements of strains or substrates) allow for the continuous increase of the productivity of bioreactors to such an extent that there is hardly any need for new bioreactors. The increase in productivity can often keep up with the increase in demand of the product. If the demand does not increase, the increased productivity often results in the availability of one or more of the existing reactors for new products. Often the main necessary changes in hardware are adaptations to the needs of the new strains (cooling and aeration capacity) to extend the "limits of growth".

When a company has decided to produce a new product in a new bioreactor the design of this reactor is often caught up by strain improvement: when the reactor is ready the strain is, as a result of ongoing improvements, already much more productive than the one used for the design calculations.

Also the quality of the substrate can vary enormously as a function of time (fluctuations due to changing seasons, geographical origin, or supplier).

The need for methods to improve the output of bioreactors increases when the product becomes a commodity and/or when it is at the end of its life cycle (cost cutting). The cost/benefit ratio for R&D for this purpose must be checked carefully and frequently (small margins).