

## Second Edition BIOCHEMICAL ENGINEERING

PRINCIPLES AND CONCEPTS

**Syed Tanveer Ahmed Inamdar** 





## **Contents**

Fo	rewor	d	$x_1$
Pr	eface		xvi
$P_T$	eface	to the First Edition	xi
	•	nical Engineering: A Perspective	xx
l.		CROBIOLOGY FUNDAMENTALS	<b>-2</b> 7
	1.1	- \ 1	
		Place of Microorganisms in the Living World 3	
		Whittaker's Five-Kingdom Concept 3	
	1.4	Structure of Cells 4	
		1.4.1 Prokaryotic Cells 4	
		1.4.2 Eukaryotic Cells 5	~
		1.4.3 Comparison between Prokaryotic and Eukaryotic Cells	7
	_		
	0.1	Cell Types (Commercially Viable) 9	
		1.6.1 Protists 9 1.6.2 Bacteria 10	
		1.6.3 Yeasts 13	
		1.6.4 Moulds 15	
		1.6.5 Algae, Protozoa and Viruses 16	
		1.6.6 Animal and Plant Cells 17	
	17	Environmental and Industrial Microbiology 17	
	1.1	1.7.1 Microbiology of Soil 18	
		1.7.2 Aquatic Microbiology 18	
		1.7.3 Domestic Water and Wastewater Microbiology 19	
		1.7.4 Microbiology of Foods 19	
	1.8	Cycles of Life 20	
	_	1.8.1 Oxygen Cycle 20	
		1.8.2 Carbon Cycle 21	
		1.8.3 Sulphur Cycle 22	
		1.8.4 Phosphorus Cycle 23	
		1.8.5 Nitrogen Cycle 24	
		1.8.6 Calcium Cycle 25	
	$\operatorname{Sun}$	nmary 26	
		rcises 26	

2.	BIC	DLOGICAL POLYMERS28-62
	2.1	Introduction to Biological Polymers 28
		2.1.1 Repetitive Biological Polymers 28
	0.0	2.1.2 Non-repetitive Biological Polymers 29
	2.2	Lipids 29
		2.2.1 Classification of Lipids $29$ 2.2.2 Functions of Lipids $3\theta$
		2.2.3 Amphipathic Lipids 30
		2.2.4 Stable Configurations of Fatty Acids in Water 31
		2.2.5 Vitamins, Steroids and Other Lipids 32
	2.3	Proteins 33
		2.3.1 Functions of Proteins 33
		2.3.2 Elemental Composition of Proteins 34
		2.3.3 Classification of Proteins 34
	2.4	Amino Acids 36
		2.4.1 Optical Isomers of Amino Acids 36
	0.5	2.4.2 Properties of Amino Acids 36
	2.5	Protein Structures 39
		2.5.1 Primary Structure of Protein 40 2.5.2 Secondary Structure of Protein 40
		2.5.3 Tertiary Structure of Protein 41
		2.5.4 Quaternary Structure of Protein 42
	2.6	Protein Denaturation and Renaturation 42
	2.7	Nucleic Acids 43
		2.7.1 DNA Structure 45
		2.7.2 Watson and Crick Model of DNA 46
		2.7.3 Ribonucleic Acid (RNA) 48
	0.0	2.7.4 Biological Information Storage and DNA Replication 49
	2.8	Carbohydrates $52$ 2.8.1 Monosaccharides $52$
		2.8.2 Disaccharides 54
		2.8.3 Polysaccharides 55
	2.9	Macronutrients and Micronutrients 58
		Hybrid Biochemicals 58
		2.10.1 Hierarchy of the Biological Structure 59
		nmary 60
	Exe	rcises 61
9	TO N.T.	ZVANEC AND ENGINAR VINERICG
3.	3.1	ZYMES AND ENZYME KINETICS 63-124  Nomenclature and Classification 64
	5.1	Nomenclature and Classification 64 3.1.1 Chemical Nature and Properties of Enzymes 64
		3.1.2 Enzymes and Their Actions 65
	3.2	Enzyme Kinetics 65
		3.2.1 Enzyme Catalysis 65
		3.2.2 Order of Reaction 66
		3.2.3 Progress of a Reaction 69
	3.3	Mechanism and Kinetics of Enzymatic Reactions 70
		3.3.1 Lock and Key Model or Fischer's Template Theory 70
		3.3.2 Induced Fit Theory or Koshland's Model 71
		3.3.3 Substrate Strain Theory 71
		3.3.4 Integral Michaelis-Menten Equation 75

3.4 Evaluation of Kinetic Parameters 78
3.4.1 The Lineweaver–Burk Method 78
3.4.2 The Eadie-Hofstee Method 79
3.4.3 The Hanes-Woolf Method 80
***** F
3.5.2 Temperature Effects 85
3.5.3 Concentration Effects $86$
3.5.4 Activators 86
3.5.5 Product Concentration 87
3.5.6 Time 87
3.5.7 Radiations 87
3.6 Features of Active Sites 87
3.7 Inhibitors and Inhibition Kinetics 87
3.7.1 Reversible Inhibition 88
3.7.2 Substrate Inhibition 96
3.7.3 Irreversible Inhibition $100$
3.7.4 Allosteric Inhibition $100$
3.7.5 Product Inhibition 102
3.7.6 The Inhibitor Constant $K_I = 102$
3.8 Enzyme Deactivation 103
3.8.1 Enzyme Deactivation Model 103
3.8.2 Combination of Deactivation Model with Simple
Catalytic Sequence of Michaelis-Menten Equation 104
3.8.3 Half-life Period 106
3.9 Kinetics of Multisubstrate Systems 106
3.9.1 The Ordered Sequence Mechanism 108
3.9.2 The Random Sequence Mechanism 109
3.9.3 The Ping-Pong or Double Displacement Mechanism 110
3.10 Modulation and Regulation of Enzyme Activity 111
3.10.1 Enzyme Regulation Mechanisms 112
3.10.2 Regulation of Enzyme Activity 113
Summary 116 Exercises 116
Exercises 116
INDUSTRIAL ENZYMES AND APPLICATIONS 125-135
4.1 Hydrolytic Enzymes 126
4.2 Proteolytic Enzymes 127
4.2.1 Methods of Activation 127
4.2.2 Applications of Proteolytic Enzymes 128
4.3 Non-hydrolytic Enzymes 129
4.3.1 Recent Applications of Non-hydrolytic Enzymes 129
4.4 Enzymes of Industrial Importance 130
4.5 Enzyme Market 131
4.6 Production of Enzymes on a Commercial Scale 131
4.6.1 Isolation of Microorganisms, Strain Development
and Preparation of Inoculum 131
4.6.3 Sterilization and Inoculation of Medium, Maintenance of
Culture and Fluid Filtration 132
4.6.4 Enzyme Purification 133
Summary 134
Exercises 135

4.

<b>5</b> .		MOBILIZED-ENZYME TECHNOLOGY136–168
	5.1	Enzyme Stabilization 136
	5.2	Immobilization 137
		5.2.1 Adsorption Immobilization 137
	5.3	
		5.3.1 Chemical Methods 139
		5.3.2 Physical Methods 140
	5.4	Diffusional Mechanisms and Limitations in the
		Immobilized Enzyme Systems 142
		5.4.1 Characterization of Immobilized Enzymatic
		Reaction Rates Using Damköhler Number 143
		5.4.2 Mechanism of Mass Transfer and Chemical Reactions
		in a Symmetric Slab of Immobilized Enzyme 144
		5.4.3 Effects of Diffusion in Surface-Bound Enzymes on a
		Non-Porous Catalyst Support 145
		5.4.4 Diffusion Effects in Enzymes Immobilized in a
		Porous Matrix Support 150
		5.4.5 Electrostatic and Position Effects in Immobilized
		Enzyme Systems 154
	5.5	Immobilized Cell Systems 154
		5.5.1 Formulation and Characterization of Immobilized
		Cell Biocatalysts 156
		5.5.2 Techniques for Cell Immobilization 157
		5.5.3 Transport Across Cell Membranes 160
	0	5.5.4 Reactor Configurations for Immobilized Cells 164
	Sum	nmary 167 rcises 168
	Exe	rcises 108
6.	BIO	MASS PRODUCTION IN CELL CULTURES 169-23;
٠.	6.1	Biomass and Cell Cultures 169
		6.1.1 Environmental Medium 171
		6.1.2 Cell Population 172
	6.2	
	6.3	Ideal Reactors for Measurement of Kinetics 173
		6.3.1 The Ideal Batch Reactor 174
		6.3.2 The Ideal Continuously Stirred Tank Reactor (CSTR) 17.
	6.4	
	6.5	Monod's Growth Kinetics 178
	6.6	Transient Growth Kinetics 179
		6.6.1 Growth Patterns and Kinetics in a Batch Culture 179
	6.7	Cell Culture Medium 182
		Environmental Factors Affecting Growth Kinetics 183
	6.9	Cell Growth and Kinetic Patterns 184
		6.9.1 Kinetic Patterns of Growth and Product Formation
		in Batch Fermentation 185
		6.9.2 Yield Coefficient and Maintenance Coefficient 187
		6.9.3 Mass Balance on Substrates 188
		6.9.4 Products Balance 190
		6.9.5 Other Forms of Growth Kinetics 190
		6.9.6 Estimation of Lag Time 190
		6.9.7 Relation between Initial Nutrient Concentration and
		Maximum Cell Population in Batch Culture 191
		6.9.8 Pearl and Reed Model (Unstructured) 191

	6.10 Reactors and Their Configurations 192 6.10.1 Growth of Microorganisms (MOs) in a Batch Reactor 193 6.10.2 Continuous Culture of Microorganisms (MOs) 195 6.10.3 Stirred Tank Reactor with Recycle of Biomass 202 6.10.4 Continuous Stirred Tank Fermenters (CSTFs) in Series 207 6.10.5 Stirred Tank Fermenters in Series (Considering N no. of Reactors) 211
	6.10.6 Stirred Tank Fermenter in Series with Biomass Recycle 213 6.10.7 Plug Flow Fermenters (PFR/PFF) 215 6.10.8 Plug Flow Fermenter (PFF) with Biomass Recycle 217 6.10.9 Evaluation of Kinetic Parameters 219
	6.10.10 Use of Unstructured, Unsegregated Models to Predict Specific Growth Rate 225 6.10.11 Models for the Growth of Filamentous Organisms 228 6.10.12 Models for Transient Behaviour 229 Summary 232 Exercises 233
7.	BIOLOGICAL REACTORS
8.	FERMENTATION TECHNOLOGY—TRADITIONAL PROCESSES AND PRODUCTS
9.	Exercises 269  DOWNSTREAM PROCESSING

	9.3	Cell Disruption 284
		9.3.1 Physical Methods 285
		9.3.2 Chemical and Enzymatic Methods 285
		9.3.3 Mechanical Methods 286
	9.4	Primary Isolation 286
		9.4.1 Solvent Extraction 287
		9.4.2 Supercritical Fluid Extraction 291
		9.4.3 Sorption 293
		9.4.4 Precipitation 294
	9.5	Purification 295
		9.5.1 Fractional Precipitation 295
		9.5.2 Adsorption Process 295
		9.5.3 Chromatography 299
	9.6	Final Product Isolation 309
		9.6.1 Freeze Drying 310
		9.6.2 Formulation <i>310</i>
	9.7	Membrane Separation Processes 311
		9.7.1 Reverse Osmosis 321
		9.7.2 Ultra-filtration 326
		9.7.3 Micro-filtration 330
		9.7.4 Dialysis 331
		9.7.5 Nano-filtration 332
	• •	9.7.6 Electrophoresis 332
		Case Studies 336
		mary 341
	HVO	
	LAC	rcises 342
10		
10.	TH	E CONTROL OF MICROORGANISMS 344–359
10.	<b>TH</b> 1	E CONTROL OF MICROORGANISMS
10.	<b>TH</b> 1110.1110.2	E CONTROL OF MICROORGANISMS
10.	THI 10.1 10.2 10.3	E CONTROL OF MICROORGANISMS
10.	THI 10.1 10.2 10.3	E CONTROL OF MICROORGANISMS
10.	THI 10.1 10.2 10.3	E CONTROL OF MICROORGANISMS
10.	THI 10.1 10.2 10.3	E CONTROL OF MICROORGANISMS
10.	THI 10.1 10.2 10.3	E CONTROL OF MICROORGANISMS
10.	THI 10.1 10.2 10.3	E CONTROL OF MICROORGANISMS
10.	THI 10.1 10.2 10.3	E CONTROL OF MICROORGANISMS
10.	THI 10.1 10.2 10.3 10.4	E CONTROL OF MICROORGANISMS
10.	THI 10.1 10.2 10.3 10.4	E CONTROL OF MICROORGANISMS
10.	THI 10.1 10.2 10.3 10.4	E CONTROL OF MICROORGANISMS
10.	THI 10.1 10.2 10.3 10.4	E CONTROL OF MICROORGANISMS
10.	THI 10.1 10.2 10.3 10.4	E CONTROL OF MICROORGANISMS
10.	THI 10.1 10.2 10.3 10.4	E CONTROL OF MICROORGANISMS
10.	THI 10.1 10.2 10.3 10.4	E CONTROL OF MICROORGANISMS
10.	THI 10.1 10.2 10.3 10.4	E CONTROL OF MICROORGANISMS
10.	THI 10.1 10.2 10.3 10.4	E CONTROL OF MICROORGANISMS
10.	10.5 10.6 10.6 10.6	E CONTROL OF MICROORGANISMS
10.	10.5 10.6 10.6 10.6 10.6	E CONTROL OF MICROORGANISMS

Appendix	A Process Kinetics and Reactor Analysis 360-369
	eactor Fundamentals 360
	eaction Rates 360
	eaction Order 361
	.3.1 Zero Order Reactions 361
	3.2 First Order Reactions 362
	3.3 Second Order Reactions 364
	eactor Analysis 365
	.4.1 Completely Mixed Batch Reactor 365
	.4.2 Continuous Flow Stirred Tank Reactors 367
Appendix	B Bioenergetics 370-380
	Ietabolism $370$
	atabolism 371
В	.2.1 Anaerobic Oxidation of Pyruvic
_	Acid (Fermentation) 372
$\mathbf{B}$	.2.2 Aerobic Oxidation of Pyruvic Acid (Krebs Cycle) 373
	.2.3 HMP Pathway 374
	nabolism 375
	folecular Genetics and Recombinant DNA Technology 375
	375 A.1 The Human Genome
	NA Analysis 376
B	5.5.1 DNA Hybridization 376
B	5.5.2 DNA Amplification 377
D.C. M	5.5.3 DNA Sequencing 379
B.0 M	Ionoclonal Antibodies (MABs) 379 3.6.1 Applications of MABs 380
Б	.0.1 Applications of MADS 500
	C Concepts in Environmental Microbiology 381-394
	acterial Nomenclature 381
_	3.1.1 Rickettsias 381
	382 Mycoplasmas 382
	3.1.3 Archaeobacteria 383
	Characteristics of Microorganisms 384 Classification of Microorganisms 385
	dentification of Microorganisms 386
	Ture Cultures and Cultural Characteristics 387  Maintenance and Preservation of Cultures 388
	Influence and Preservation of Cultures 388  Iunicipal Water Purification 389
	Vastewater Treatment 390
	ficrobial Interactions 392
	Siodegradation 393
	Bioleaching or Microbial Leaching 394
_	
Bibliograp	phy
Index	