

CONTENTS

PART 1: STRUCTURE AND FUNCTION OF ENZYMES

Chapter 1 An Introduction to Enzymes

1.1	What are Enzymes?	17
1.2	A Brief History of Enzymes	18
1.3	The Naming and Classification of Enzymes	18

Chapter 2 The Structure of proteins

2.1	Introduction	29
2.2	Amino-Acids, The Building Blocks of Proteins	30
2.3	The Basis of Protein Structure	33
2.4	The Determination of Primary Structure	37
2.5	The Determination of Protein Structure by X-ray Crystallography	46
2.6	The Investigation of Protein Structure in Solution	56

Chapter 3 The Biosynthesis and Properties of Proteins

3.1	The Biosynthesis of Proteins	59
3.2	The Properties of Proteins	69

Chapter 4 Specificity of Enzyme Action

4.1	Types of Specificity	79
4.2	The Active Site	80
4.3	The Fischer Lock-and-Key Hypothesis	82
4.4	The Koshland Induced-Fit Hypothesis	82
4.5	Hypothesis Involving Strain or Transition-State Stabilisation	85
4.6	Further Comments on Specificity	86

Chapter 5 Monomeric and Oligomeric Enzymes

5.1	Monomeric Enzymes	88
5.2	Oligomeric Enzymes	91

PART 2: KINETIC AND CHEMICAL MECHANISMS OF ENZYME-CATALYSED REACTIONS

Chapter 6 An Introduction to Bioenergetics, Catalysis and Kinetics

6.1	Some Concepts of Bioenergetics	97
6.2	Factors Affecting the Rates of Chemical Reactions	102
6.3	Kinetics of Uncatalysed Chemical Reactions	106
6.4	Kinetics of Enzyme-Catalysed Reactions: An Historical Introduction	109
6.5	Methods Used for Investigating the Kinetics of Enzyme-Catalysed Reactions	111
6.6	The Nature of Enzyme Catalysis	114

Chapter 7 Kinetics of Single-Substrate Enzyme-Catalysed Reactions

7.1	The Relationship Between Initial Velocity and Substrate Concentration	119
7.2	Rapid-Reaction Kinetics	131

Chapter 8 Enzyme Inhibition

8.1	Introduction	142
8.2	Reversible Inhibition	142
8.3	Irreversible Inhibition	164

Chapter 9 Kinetics of Multi-Substrate Enzyme-Catalysed Reactions

9.1	Examples of Possible Mechanisms	170
9.2	Steady-State Kinetics	172
9.3	Investigation of Reaction Mechanisms Using Steady –State Methods	177
9.4	Investigation of Reaction Mechanisms Using Non-Steady-State Methods	183

Chapter 10 The Investigation of Active Site Structure

10.1	The Identification of Binding Sites and Catalytic Sites	191
10.2	The Investigation of the Three-Dimensional Structures of Active Sites	203

Chapter 11 The Chemical Nature of Enzyme Catalysis

11.1	An Introduction to Reaction Mechanisms in Organic Chemistry	208
11.2	Mechanisms of Catalysis	210
11.3	Mechanisms of Reactions Catalysed by Enzymes without Cofactors	213
11.4	Metal-Activated Enzymes and Metalloenzymes	219
11.5	The Involvement of Coenzymes in Enzyme-Catalysed Reactions	224

Chapter 12 The Binding of Ligands to Proteins	
12.1 Introduction	239
12.2 The Binding of a Ligand to a Protein Having a Single Ligand-Binding Site	239
12.3 Cooperativity	240
12.4 Positive Homotropic Cooperativity and Hill Equation	241
12.5 The Adair Equation for the Binding of a Ligand to a Protein having Two Binding Sites for that Ligand	244
12.6 The Adair Equation for the Binding of a Ligand to a Protein having Three Binding Sites for that Ligand	248
12.7 The Adair Equation for the Binding of a Ligand to a Protein having Four Binding Sites for that Ligand	249
12.8 Investigation of Cooperative Effects	250
12.9 The Binding of Oxygen to Haemoglobin	253

Chapter 13 Sigmoidal Kinetics and Allosteric Enzymes

13.1 Introduction	257
13.2 The Monod-Wyman-Changeux (MWC) Model	257
13.3 The Koshland-Nemethy-Filmer (KNF) Model	264
13.4 Differentiation Between Models for Cooperative Binding in Proteins	267
13.5 Sigmoidal Kinetics in the Absence of Cooperative Binding	268

Chapter 14 The Significance of Sigmoidal Behaviour

14.1 The Physiological Importance of Cooperative Oxygen-Binding by Haemoglobin	275
14.2 Allosteric Enzymes and Metabolic Regulation	277

PART 3: APPLICATION OF ENZYMOLOGY

Chapter 15 Investigation of Enzymes in Biological Preparations

15.1 Choice of Preparation for the Investigation of Enzyme Characteristics	294
15.2 Enzyme Assay	296
15.3 Investigation of Sub-Cellular Compartmentation of Enzymes	305

Chapter 16 Extraction and Purification of Enzymes

16.1 Extraction of Enzymes	314
16.2 Purification of Enzymes	320

Chapter 17 Enzymes as Analytical Reagents

17.1 The Value of Enzymes as Analytical Reagents	332
17.2 Principles of Enzymatic Analysis	333

Chapter 18 Instrumental Techniques Available for Use in Enzymatic Analysis	
18.1 Principles of the Available Detection Techniques	345
18.2 Automation in Enzymatic Analysis	354
Chapter 19 Some Applications of Enzymatic Analysis in Medicine and Industry	
19.1 Applications in Medicine	363
19.2 Applications in Industry	373
Chapter 20 Biotechnological Applications of Enzymes	
20.1 Large-Scale Production of Enzymes	376
20.2 Immobilised Enzymes	379
20.3 Enzyme Utilisation in Industry	387
Answers to Problems	393
Abbreviations	398
Index	399