(L:T:P = 4:0:0:) 100 Marks

III-SEMESTER

OET 3.1: FUNDAMENTALS OF ENZYMOLOGY. (4 CREDITS: 64 h)

Unit-I, 1 credit, 16 h

Introduction to Enzymes: History, General characteristics, Nomenclature and Classification of enzymes. Catalytic power, specificity and active site. IUB enzyme classification. Definition of IU, Ketal, enzyme turnover number and specific activity.

Enzyme purification: Enzyme localization, isolation and purification of enzymes. Methods employed in isolation, purification and characterization of enzymes. Presentation of enzyme purification data and criteria of purity of enzymes. Fundamentals of enzyme assay. Assay of enzymes by Spectrometric,colorimetric, manometric, fluorimetric, isotopic methods.

Unit-II, 1 credit, 16 h

Kinetics of enzyme- Catalyzed reactions:

Enzyme kinetics of single substrate reactions- Michaelis theory, steady state theory. Kinetic data evaluation- linear transformation of Michaelis-Menten equation. Pre-steady state kinetics. Integrated velocity equation, King-Altman procedure for deriving the rate equation. Effect of pH and temperature. Enzyme Inhibition- reversible; competitive, non-competitive, uncompetitive, irreversible inhibition.

Distinction between different kinetic pathways using primary and secondary plots. Inhibition studies.

Unit-III , 1 credit, 16 h

Chemical nature of enzyme catalysis- General acid-base catalysis, electrostatic catalysis, covalent catalysis, intramolecular catalysis and enzyme-catalysis.

The investigation of active site structure: The identification of binding sites and catalytic sites- trapping the E-S complex- the use of substrate analogs, chemical modification of amino acid side chains, photo- oxidation, enzyme modification by treatment with proteolytic enzymes.

Mechanisms of reactions catalyzed by the following enzymes- Chymotrypsin, and Ribonuclease.

Coenzymes: The mechanistic role of the following coenzymes in enzyme catalyzed reactions- nicotinamide nucleotides, flavin nucleotides, pyridoxal phosphate, coenzyme A, lipoic acid, thiamine pyrophosphate, biotin,tetrahydrofolate.

Unit-IV , 1 credit, 16 h

Monomeric, oligomeric and multienzyme complexes; The serine proteases and alcohol dehydrogenase. Isoenzymes- Lactate dehydrogenase (LDH) , Pyruvate dehydrogenase (PDH) complex.

Regulation of activity of enzymes: Reversible and irreversible covalent modifications, allosteric modifications.

Applications of enzymes: Industrial, Clinical, Analytical and Biotechnological applications.Immobilized enzymes: Principle, advantages and limitation, methods of enzymes immobilizations- Adsorption, flocculates, chemical bonding- cross-linking, gelatin encapsulation. Applications of immobilized enzymes.

REFERENCES

1 Basic Biochemical Laboratory Procedures and Computing. R. Cecil Jack. Oxford University.
2 Protein Purification Methods. S.L.V. Harris and Angal. IRL Press.
3 Understanding Enzymes. Palmer. T. Ellis Horwood Ltd.
4 Enzyme Kinetics. Roberts. D.V. Cambridge University Press.
5 The Enzymes. Boyer. Academic Press.
6 Enzyme Kinetics. Irwin H. Segel. Interscience-Wiley.
7 Enzyme Kinetics:The Steady state approach. Engel, P.C. 2nd Edn. Champman and Hall.
8 Nature of Enzymology. Foster.Croom Helm.
9 Principles of Enzymology for Food Sciences. Whitaker, Marcel Dekker. Academic Press.
10 Fundamentals of Enzymology. N.C. Price and Lewis. Oxford University Press.

OEP3.1: Practicals based on OET3.1
(L:T:P = 0:0:2) 50 Marks