## **Enzymes Biotechnology Handbook**

Author:- NIIR Board of Consultants and Engineers Format: paperback Code: NI98 Pages: 490 Price: Rs.1100US\$ 125 Publisher: NIIR PROJECT CONSULTANCY SERVICES Usually ships within 5 days

Industrial biotechnology is the practice of using cells to generate industrially useful products. An enzyme is a protein that catalyzes, or speeds up, a chemical reaction. Enzymes are the focal point of biotechnological processes, without them biotechnology as a subject would not exist. The main advantage of enzymes compared to most other catalysts is their stereo, region and chemo selectivity and specificity. Enzymes are responsible for many essential biochemical reactions in micro organisms, plants, animals, and human beings. Biotechnology processes may have potential in energy production, specifically in the substitution of renewable plant biomass for fossil feedstock. This will depend on the development of enzymes able to degrade cellulose in plant biomass and designing methods to recycle or dispose of spent biomass. With time, research, and improved protein engineering methods, many enzymes have been genetically modified to be more effective at the desired temperatures, pH, or under other manufacturing conditions typically inhibitory to enzyme activity (e.g. harsh chemicals), making them more suitable and efficient for industrial or home applications. Enzymes are used in the extraction of natural products, as catalysts in organic chemistry, in clinical analysis, in industrial processes, and so on. The application of enzymes is found in many different fields and it is one of the good sectors to venture. In coming few years it is estimated that world enzyme demand will average annual increases of 6.3 percent.

This book basically deals with principles of industrial enzymology, basis of utilization of soluble and immobilized, enzymes in industrial processes, principles of immobilization of enzymes, enzymes in clinical analysis principles, practical aspects of large-scale protein purification, the applications of enzymes in industry, use of enzymes in the extraction of natural products, data on techniques of enzyme immobilization and bio affinity procedures etc. In this book you can find all the basic information required on the fundamental aspects of the enzymes, their chemistry, bio chemistry as well as detailed information of their applications a wide variety of industrial processes etc. The book is very useful for research scholars, technocrats, institutional libraries and entrepreneurs who want to enter into the field of manufacturing of enzymes.

1. LARGE-SCALE EXTRACTION AND PURIFICATION OF ENZYMES AND OTHER PROTEINS Extraction by Chemical Methods Alkali Lysozyme and EDTA Detergents Cold Shock Osmotic Shock **Extraction by Physical Methods** Sonication Freezing and Thawing Solid Shear Grinding or Agitation with Abrasives Liquid Shear **Isolation and Purification** Nucleic Acid Removal **Cetyltrimethyl Ammonium Bromide** Streptomycin Sulphate Polyethyleneimine Nuclease Treatment Concentration by Precipitation Ammonium Sulphate **Organic Solvents High Molecular Weight Polymers** Concentration by Ultrafiltration Concentration by Freeze-Drying Gel Chromatography Ion Exchange Chromatography Ion Exchange Resins Ion Echange Celluloses Other Ion Exchange Gels Affinity Chromatography **Coupling Techniques** Non-Specific Adsorbents Hydroxyapatite Celite Hydrophobic Interaction Chromatography High Performance Liquid Chromatography **Electrophoretic Techniques** Electrophoresis. **Isoelectric Focusing** Multimembrane Electrodecantation Chromatofocusing Aqueous Two-Phase Separation 2. PRINCIPLES OF INDUSTRIAL ENZYMOLOGY: BASIS OF UTZATION OF SOLUBLE AND **IMMOBILIZED** ENZYMES IN INDUSTRIAL PROCESSES Glossary of Symbols Assay of Enzyme Activity Cofactors The Distinctive Features of Enzymes as Catalysts **Enzyme Catalysis Enzyme Kinetics** The Effect of pH on Enzyme Activity The Effect of Temperature on Enzyme Activity **Enzyme Inhibition** The Various Types of Enzymic Catalyst A Comparison of Enzymes with Chemical Catalysts A Comparison of Enzymes with Fermentations

Immobilized Biocatalysts A Comparison of Immobilized Enzymes and Cells An Assessment of Immobilization Supports and Methods Characterisation of Immobilized Biocatalysts **Co-Immobilized Enzymes Two-Phase Reaction** Industrial Enzyme Kinetic Effects on Equilibria **Effectiveness Factors Steady - State Kinetics** Intrinsic Activity of Enzymes - Modifying Factors Introduction Diffusional Limitations on the Activity of Immobilized **Biocatalysts External Diffusional Limitations** Internal Diffusional Restrictions **Regeneration of Cofactors Biochemical Reactors** Introduction The Various Types of Biochemical Reactor Assessment of the Performance of Biochemical Reactors **Batch Reactors Continuous Stirred Tank Reactors** Plug-flow Column Reactors (or Tubular Reactors) Fluidized Bed Reactors **Electrochemical Reactors** Ultrafiltration Reactors **Enzyme Kinetic in Reactors** Inhibition in Enzyme Reactors The Effect of Non-Ideal Flow on Biochemical **Reactor Performance** Physical Problems in Biochemical Reactor using Immobilized Biocatalysts Abrasion Compression Fouling **Microbial Contamination** The Stability of Immobilized Biocatalysts Introduction The Stability of Biochemical Reactors Employing Immobilized Enzymes or Immobilized Cells **Regeneration of Biocatalyst Activities** Constant Productivity with Biocatalyst Reactors Scale-Up Discussion 3. PRINCIPLES OF IMMOBILIZATION OF ENZYMES **Classification of Immobilized Enzymes** Techniques of Enzyme Immobilization Entrapment Gel Entrapment Fibre Entrapment Micro-encapsulation **Carrier Binding** 

**Physical Adsorption** Ionic Binding Method Chelation or Metal Binding **Covalent Binding** Crosslinking Immobilized Soluble Enzymes Immobilization without Enzyme Derivitization Immobilization with Enzyme Derivitization **Miscellaneous Methods** Choice of Immobilization Method Outline of Properties of Immobilized Enzymes Stability **Kinetic Properties Outline of Enzyme Reactors Batch Reactors Continuous Reactors Application and Future Trends Analytical Applications Enzyme Electrodes** Automated Analysis **Therapeutic Applications Enzyme Replacement** Enzyme Therapy Industrial Applications Future Trends 4. ENZYMES IN CLINICAL ANALYSIS - PRINCIPLES Measurment of Substrate Concentration with Enzymes Principles of Equilibrium Methods Principles of Kinetic Methods Comparison of Equilibrium and Kinetic Methods Common Indicator Species Used in Routine Clinical Analysis Nicotinamide Adenine Dinucleotides Oxygen and Hydrogen Peroxide Measurement of Enzymes Principles of Enzyme Assay Using Coupled Enzymes Immobilized Enzymes for Measuring Substrate Concentrations Immobilized Enzyme Reactor Tubes **Bioanalytical Probes** Dry Reagent Chemistry Enzyme Immunoassay (ELA) Preparation of Enzyme Labels Homogeneous EIA Heterogeneous EIA Choice of Enzyme Label Assay in EIA Simultaneous Assay of Two Haptens The Future 5. PRACTICAL ASPECTS OF LARGE-SCALE PROTEIN PURIFICATION **Enzyme Inactivation Containers and Ancillary Equipment Glass Vessels** 

Metal Vessels Plastic Vessels Liouid Transer Couplings Pumps **Bacterial Disruption** Resuspension Liquid Shear Grinding Centrifugation **Batch Centrifuges Continuous Flow Centrifuges Disc Type Centrifuge** Hollow Bowl Centrifuges **Basket Centrifuges Tangential Flow Filration** Concentration Ultrafiltration Stirred Cells Thin Channel Systems Cartridge Membranes Hollow Fibres Dialysis Chromatography 1 Columns Gel Chromatography Ion Exchange Chromatography Affinity Chromatography 6. THE APPLICATIONS OF ENZYMES IN INDUSTRY Glossarv of Terms **Production of Enzymes** Use of Enzyme - General Comments The Characteristics of Industrial Enzymes Soures of Enzymes The Isolation, Purification and Formulation of Enzymes Legislation on the Use of Enzymes **Enzyme Manufacturers Biochemical Applications** Use of Enzymes in Analysis General In Clinical Assays Medical Uses of Enzymes The Use of Enzymes as Catalysts in Organic Chemistry Introduction Stereospecificity of Enzymes **Prochiral Stereospecificity Combinations of Stereospecificity Multiple-Step Reactions** Synthesis of Radioactive Compounds **Restrication Endomucleases Biochemical Processing** Applications of Enzymes in the Food Industry Polysaccharide Processing

**Bacterial** -amylase Amyglucosidase (EC 3.2.1.3) Maltose Syrups Glucose Isomerase (EC 53.1.5) Inversion of Sucrose Sugar Refining Raffinase a-amylase Dextranase (EC3.2.1.11) **Debranching Enzymes** Cyclodextrin Glucosyltransferase and Other Amylases Cellulase (EC3.2.1.4) **Ethanol Fermentation** Brewing Baking The Dairy Industry Lactose Hydrolysis Cheese Manufacture Coagulation Flavour Development Other Applications **Organic Acids** Amino Acids Introduction **Enzymic Resolution Enzymic Production of Amino Acids** Antioxident Introduction Glucose Oxidase (EC 1.1.3.4) **Protein Processing** Introduction The Plastein Reaction Aspartame Others **Flavouring Agents** Fruit Processing Use of Enzymes in the Extraction of Natural Products **Detoxifying Enzymes Enzyme-Based Detergents** Use of Enzymes as Cleansing Agents The Leather Industry Textiles Paper Manufacture Antibiotics Penicillin Acylase, (EC 3.5.1.11) Cephalosporins Miscellaneous uses of Biocatalysts Conclusion Note in Proff Acrylamide **Propylene Oxide** Vinyl Chloride **Biosensors** 

Amino Acids 7. DATA ON TECHNIQUES OF ENZYME IMMOBILIZATION AND BIOAFFINITY PROCEDURES Entrapment Gel Entrapment **Fibre Entrapment** Microencapsulation Phase Separation Method Interfacial Polymerization Method Liquid Surfactant Membrane Method Liquid Drying Method Chelation or Metal Binding **Covalent Binding** Diazotization **Amide Bond Formation** Acid Anhydride Derivatives Acylazide Derivatives Cyclic Imidocarbonate Derivatives Isocyanatc and Isothiocyanate Derivatives Acyl Chloride Derivatives Cyclic Carbonate Derivatives **Condensing Reagents** Alkylation and Arylation Schiff's Base Formation Ugi Reaction **Amidination Reactions Thiol-Disulphide Interchange** Mercury-Enzyme Interactions -Irradiation Induced Coupling Matrices for Carrier Binding **Inorganic Supports Controlled Pore Supports Other Porous Supports** Non-porous supports **Coupling Reactions for Inorganic Supports Organic Supports** Polysaccharides **Proteins** Carbon Polystyrenes Polyacrylates Maleic Anhydride Based Copolymers **Polypeptides** Vinyl and Allyl Polymers Polyamides Crosslinking **Immobilized Cells** Entrapment **Physical Adsorption** Chelation or Metal Binding **Covalent Binding** Crosslinking Other Immobilized Biologically Active Moleccules

Immunoadsorbents Affinity Chromatography Media Immobilized Lactins Immobilized Amino Ecids and Peptides Immobilized Carbohydrates Immobilized Nucleosides, Nucleotides and Nucleic Acids Immobilized Antibiotics 8. ENZYMES IN CLINICAL ANALYSES - DATA Substrates Measured Enzymically in Clinical Laboratories Enzymes Measured by Coupled Enzyme System in Clinical Laboratories Immobilized Enzymes for Measuring Substrates Enzymes Used in Enzyme Immunoassay (EIA)

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Fri, 02 May 2025 16:38:55 +0000