PREFACE

Biotechnology has emerged as an area of activity having tremendous impact on virtually all domains of human welfare and holds a lot of promises and surprises in some of the techniques for the present and future. Spread over fourteen chapters, the book gives an overview of biotechnological means to convert hazardous wastes into useful products, then moves to an important area of biotechnology i.e. proteomics. It touches upon the latest research and developments in diverse areas like bio-informatics, DNA bio-sensors, DNA fingerprinting, mushroom biotechnology, stem cells, bioethics, in vivo induced antigen technology, immobilization technology, production of 7- aminocephalosporanic acid, therapeutic application of thymic peptides and cyanobacteria as a source of valuable products.

Chapter 1 - Management of voluminous quantities of hazardous wastes by physical, chemical and physicochemical means has proved to be inefficient, uneconomical and unecofriendly. On the other hand biotechnology which uses the biological processes has great potential to degrade the wastes into products that are part of naturally occurring cycles. Biotechnology includes a range of technologies which are applied in the purification of polluted air, treatment of wastewater and cleaning of contaminated soil. The bioremediation and phytoremediation techniques which use the microbial organisms and plants, respectively to detoxify hazardous wastes has received global attention as effective biotechnological approach to clean up the environment. Recombinant DNA technique has made it possible to create microorganisms with desired genes and subsequently use of these microorganisms in detoxification of hazardous wastes.

Chapter 2 - Proteomics involves the systemic study of proteins in order to provide a comprehensive view of the structure, function and regulation of biological systems. Advances in instrumentation and methodologies have fueled an expansion of the scope of biological studies from simple biochemical analysis of single proteins to measurement of complex protein mixtures. Proteomics is rapidly becoming an essential component of biological research. Coupled with advances in bioinformatics, this approach to comprehensively describing biological systems will undoubtedly have a major impact on our understanding of the phenotypes of both normal and diseased cells. Initially proteomics focused on the generation of protein maps using two-dimensional polyacrylamide gel electrophoresis. The field has since expanded to include not only protein expression profiling, but also the analysis of post-translational modifications and protein-protein interactions. However, mass spectroscopy has been incorporated to increase sensitivity, specificity and to provide results

in a high throughput format. The study of protein-protein interactions has been revolutionized by the development of protein microarrays.

Chapter 3 - The development of databases and the use of bioinformatics have not only made science interesting but also easier to carry out new researches using the existing biological data. Enormous amount of the available data could be sorted out with ease as per the demand of the topic of the research using various algorithms, machine learning approaches and information retrieval systems. Genomics and Proteomics are an area of research which have still many mysteries unsolved and here the approaches of bioinformatics has made the tedious work much more simpler by providing numerous popular tools like BLAST and FASTA, ClustalW, PSI-Blast etc. to sort out the aggregate features from the completely sequenced genomic data. New methodologies are being developed to identify distant homologs, presence and absence of a protein in any sequenced genome, prediction of gene functions and many more. The role of bioinformatics in determining the complex structure of proteins, *de novo* modeling of loops in protein structure is worth mentioning. The revolution of bioinformatics in drug designing has given the hope to conquer the deadly diseases. The development of novel enzyme inhibitors like HIV-1 protease has now become possible. A number of programs for prediction of various ADME (Adsorption, distribution, metabolism and excretion) properties of hit molecules are successfully designed. Various computer-assisted drug discovery programs were developed for a wide range of applications including rigid body protein ligand docking, flexible docking and protein-protein interactions.

Chapter 4 - DNA based biosensors have recently gained much importance for detection of target genes responsible for diseases, in food industry, environment and in agriculture. Different types of DNA based biosensor, their advantages and basic principle of operating system has been described in this chapter. DNA biosensor provides fast, simple, economical, sensitive and selective detection of target genes by hybridization with specific probe. Various new strategies for DNA based biosensors have been described along with recent trends and challenges in development of technology. Electrochemical biosensor has more advantages due to the hybridization reaction on electrode surface or in solution in the presence of redox indicators. PCR free DNA biochip is emerging **as** new tools in the field of diagnosis.

Chapter 5 - DNA fingerprinting is a very powerful technology which is used by forensic scientists to establish identity. Such DNA-based technologies can help in the fight against crime. This technology has experienced many advancements occurring with the passage of time. Variable number tandem repeats (VNTR) are abundant in our genome and these regions are analyzed in DNA profiling.DNA testing is a matter of serious concern as it involves many ethical issues. DNA fingerprinting can be a boon to our society in different walks of our life if used efficiently.

Chapter 6 - Mushroom biotechnology is concerned with all aspects of the application of mushroom i.e. domestication of wild variety, enhanced production of nutriceuticals, bioconversion of lignocellulosic wastes into useful products, biodegradation of recalcitrant substances and bioremediation of xenobiotics. Molecular tools are being used for the improvement of mushroom strains which in turn can act as an effective weapon against malnutrition, many diseases and environmental pollution. Gene transfer using protoplast has been developed to break down the barrier of gene imposed by conventional breeding systems. Protoplast technology provides a highly feasible technique for interspecific and intergeneric hybridization. The versatile enzymatic systems of some of the mushrooms, which attack

lignin and have low substrate specificity, are exploited to degrade many toxic and recalcitrant pollutants.

Chapter 7 - Treatment of tissue degenerative diseases like neurodegenerative disease, type one diabetes mellitus, spinal cord injury, chronic liver disorder, etc. and understanding various aspects of such diseases have remained as major challenges in the biomedical sciences. Isolation of embryonic stem (ES) cells opens up the possibility to meet these chanllanges although ethical and immunological issues in using these cells pose serious restrictions. Recent progress in developing methods to efficiently dedifferentiate cells from adult body into ES cells, known as induced pluripotent stem (iPS) cells, offers a promising and viable alternative. Use of iPS cells also bypasses ethical as well as immunological issues. IPS cell therapy, therefore, is being percied as the autogenic cell source for treatment of various tissue degenerative diseases.

Chapter 8 - Stem cells are cells found in most of the multi-cellular organisms. They are characterized by the ability to renew themselves through mitotic cell division and differentiating into a diverse range of specialized cell types. The prospect of this technology is immense despite the fact that research in the stem cell segment has been exhaustively discussed and criticized at numerous political, ethical and public forums in several countries. It has raised a hope among the scientist community about the possible use of stem cells as a tool not only to replace parts for failing organs but also to treat certain debilitating diseases like diabetes and Parkinson's disease. Most cells within an animal or human body are committed in fulfilling a specific function within the body. In contrast, stem cells are unique and important set of cells that are not specialized. These cells retain the ability to differentiate into many or all of the different cell types in the body and thereby play a critical role in repairing organs and body tissues throughout life. They can therapeutically divide without limit to replenish other cells as long as the person or animal is alive. When a stem cell divides, each new cell has the potential to either remain a stem cell or become another type of cell with a more specialized function, such as a muscle cell, red blood cell, or brain cell. Apart from application of stem cells in regenerative therapy, it also helps in understanding the early human development and the pathophysiology of genetic diseases at the cellular level. However, there are some technical problems and ethical concerns that need to be addressed before applying stem cells into clinical practice. The present chapter describes some salient features, current scenario and future prospects / retrospect of clinical applications of stem cells.

Chapter 9 - There are some common tenets to guide our actions and these are accepted as paradigm of morality in almost all human societies irrespective of differences in creed and culture. But in specific cases often unanimity is lacking. One basic criterion of morality is the acceptance of responsibility by the decision taker. New researches in the field of medical sciences are creating new problems which were hitherto not faced. New techniques in biotechnology have paved way for transplantation for kidney, heart and liver successfully. *In vitro* fertilization, surrogate mothership, researches in plelipotent stem cells which are capable of cloning are creating moral dilemma for the scientists. These techniques have immense capacity for the wellness of human beings. But there is every possibility of their misuse. Should researches in biotechnology be allowed unconditionally or there be some guiding principles? If the technique of cloning is perfected what will be the fate of the society which is based on conjugal love and trust? Should abortion and euthanasia be allowed? These questions need a clear answer. In our view no such laws can be framed which can guide

scientists in each and every case. Each case is specific and requires particular attention. There should be some norms for individual decision taker which are given in spiritual book of Hindu 'Shrimad Bhagavata Geeta'. With the help of this technique of decision making specific cases can be dealt with.

Chapter 10 - IVIAT, abbreviated for *in vivo* induced antigen technology, is one of the recently invented techniques for identification of genes of a disease causing pathogen specifically induced upon infection of host. In simpler words these genes are switched on *in vivo* but remain switched off while growing *in vitro* outside the host. Since these genes are transcribed in the host environment, it is assumed that they may be essential for infection process, survival in the host environment and progression of disease. Identification of *in vivo* specific genes may thus be important in development of chemotherapeutics, vaccines and diagnostics. IVIAT offers several advantages over other techniques developed to select out genes induced *in vivo*. IVIAT circumvents the use of animal models and can be applied to detect temporal expression of antigens through infection process.

Chapter 11 - The immobilization of enzyme and cells is a practical method when bioprocess employing biocatalysts are considered. At present, the application of immobilized biocatalyst and cells in bioprocessing include production of energy, drugs, and enzymes, valuable proteins, specialty products etc. pollutant treatment, biosensors with high sensitivity and specificity through various techniques of immobilization protocol, numerous numbers of matrix materials were tried. Recent advancement in sophisticated instrumentation and recombinant DNA technology added new dimension to this technology not only considering the biological, biochemical aspects, but also of economical factors engineering aspects such as mass transfer resistance and diffusional limitations when dealing with this technology at commercial scale.

Chapter 12 - Cephalosporin acylases are industrially important enzymes, which hydrolyze cephalosporins to 7-aminocephalosporanic acid (7-ACA), a key intermediate required for the production of most of the clinically used cephalosporin derivatives i.e. semisynthetic cephalosporins. Cephalosporin-C (CPC), an important β-lactam antibiotic in the international market is exclusively produced by aerobic fermentation as such exhibits negligible antimicrobial activity but substitutions on C_3 and C_7 positions of β-lactam ring along with other structures generate semisynthetic cephalosporins with diversified antimicrobial activity e.g. cefazolin, cefotoxime, cephamandole and cephaclor etc. The production of 7-ACA and CPC was extensively reviewed in the chapter with some of our findings using both free and immobilized microbial cells.

Chapter 13 - Thymus, a gland responsible for production of T lymphocyte has been demonstrated to be a source of a variety of short peptide hormones collectively designated as thymic peptides. These polypeptides have gained increasing clinical therapeutic applications in a wide variety of patho-physiological anomalies cancer, neuroendocrine disorders, infections, senescence and autoimmune diseases. The thymic peptides depending on their chemical composition have been categorized into different families among which thymosins are the most investigated ones. Thymosin-α and –β have been shown to participate in immunomodulation and tumor progression. We have demonstrated from studies in our laboratory that thymosinα1 can reverse tumor induced immunosuppression for therapeutic benefit of the tumor bearing host. This chapter provides a brief review of current state of

knowledge about the biological functions of thymic peptides and their potential for clinical applications and possible future directions for their therapeutic optimization.

Chapter 14 - Cyanobacteria, a small group of photosynthetic prokaryotes cosmopolitan in distribution have long been used as a model system to observe various physiological and biochemical processes. A little attention has so far been paid to the cyanobacterial secondary metabolites. The secondary metabolites not needed for normal growth exhibit antiviral, antibacterial, antifungal, antitumor, herbicidal, algicidal, antimalarial, inflammatory activities and if exploited reasonably can be of immense help to humankind. *Spirulina* is in use as human food because of its high nutritional value, high protein content (70% of dry weight) and presence of all the vitamins. Pigments especially phycobillins are in high demand in cosmetics and as food coloring agent. Cyanobacterial species are efficient catalysts of solar hydrogen production through photolysis of water. They also produce exopolysaccharides and flocculants useful in waste water treatment and clarification of solar ponds. Due to N_2 – fixing ability, cyanobacteria free – living or in symbiosis are often used as biofertilizer and reclamatization of saline usar soil in flooded paddy and sugarcane fields (specially in tropical soils). Traditional varieties of rice acquire their nitrogen need through biological sources such as cyanobacteria. *Azolla-Anabaena* association which reduces acetylene at rates that are 4-8 times greater than those observed for free-living *Anabaena cylindrica* is an efficient green manure. Cyanobacteria having simple growth needs may be exploited for the production of recombinant compounds of medicinal and commercial value as well as for the construction of mutants efficient in N_2-fixation and ammonium liberation. Cyanobacteria, unlike bacteria have been neglected as a source of valuable commercial products. This chapter emphasizes a need of multidisciplinary approach to harness the beneficial aspects related to biotechnological processes.