# BIOTECHNOLOGY AND ITS APPLICATIONS

✓ Biotechnology deals with Making genetically Modified organisMs (Microbes, plants, aniMals) for industrial production of Bio-PharMaceuticals and other useful products.

v/ Its application includes therapeutics, diagnostics, genetically Modified crops for agriculture, processed food,bio reMediation, waste treatMent and energy production. v/ The Main three critical research areas of biotechnology includes -

Providing the best catalyst in the forM ofiMproved organisM usually a microbes or pure enzyMe.

Creating optiMal conditions through engineering for a catalyst to act.

Downstream processing technologies to purify the protein or organic compounds. Biotechnological Applications in Agriculture

The three options that can be thought for increasing food production are, a) Agro-cheMical based agriculture b) Organic agriculture and c) Genetically engineered crop-based agriculture.

✓ Green revolution successfully increased the food production Many folds by using better ManageMentpractices and use of agrocheMicals, fertilizers and pesticides.

✓ But yet it was not enough to Meet the food needs the growing huMan population.

✓ Scientists have decided that use of genetically Modified crops is a possible solution.

✓ Plants, bacteria, fungi and aniMals whose genes have been altered by Manipulation are called GeneticallyModified OrganisMs (GMO).

✓ Genetic Modifications has Many advantages.

o Made crops More tolerant to abiotic stresses o Reduced reliance on cheMical pesticides o Helped to reduce post harvest losses o Increased efficiency of Mineral usage by plants o Enhanced nutritional value of food, eg., Vitamin 'A' enriched rice( Golden rice). Application of Biotechnology in production of pest-resistant plants v/ Pest resistant plants decrease the amount of pesticides used. v/ Bttoxin is produced by a bacteriuM called Bacillus thuringiensis.

v/ Bt toxin gene has been cloned froM the bacteria and been expressed in plants to provide resistance toinsects,This reduces the use of insecticides.

v/ Examples are Bt cotton, Bt corn, rice, t0Mato, potato and soya bean etc Bt cotton

✓ Bacterium Bacillus thuringiensis produce proteins that kill certain insects like lepidopterans, coleopterans(beetles) and dipterans (flies, Mosquitoes).

✓ Bacillus thuringiensis produce crystals that contain a toxic insecticidal protein.

This toxic protein present in bacterium as inactive pro toxins but as soon as insect ingest the inactive formit is converted into an active form of toxin due to the alkaline pH of the gut which solubilise the crystals.

✓ The activated toxin binds to the surface of Mid-gut epithelial cells and create pores that cause cellswelling and lysis and eventually cause death of the insect.

✓ The gene from B. thuringiensis has been incorporated into several crop plants like Cotton, Maize,

Rice etc.

✓ The toxin is coded by a gene named cry. The protein coded by the genes cry IA c and cry 11 Ab control thecotton boll worms, cry 1 Ab controls corn borers.

Pest Resistant Plants

✓ Nematode like Meloidegyne incognitia infects the roots of tobacco plants and causes reduction in yield.

✓ The infestation of these nematodes can be prevented by the process of RNA interference (RNA i). ✓ RNAi is present in all eukaryotic organisms as cellular defence by silencing of specific mRNA due to complementary dsRNA Molecules that bind to and prevents translation of the mRNA (silencing).

✓ The source of complementary dsRNA May be from an infection by viruses having RNA genomes or Mobilegenetic elements (Transposons) that replicate through RNA intermeediate. ✓ Nemaatode specific genes were introduced into host plant using Agrobacterium vectors. ✓ The introduction of DNA was such that it produced both sense and antisense RNA in the host cells.

✓ These two RNA's being complementary to each other formed a double stranded (ds DNA) that initiatedRNAi and thus, silenced the specific mRNA of the nematode.

✓ The parasite could not survive in a transgenic host expressing specific interfering RNA. Biotechnological Applications in Medicine v/ The r-DNA technological processes have made immense impact in the area of healthcare by enabling massproduction of safe and more effective therapeutic drugs.

v/ At present, about 30 recombinant therapeutics have been approved for huMan use all over the world.

1. Genetically Engineered Insulin

✓ Diabetes is caused due to the decreased production of insulin.

✓ Adult diabetes can be controlled by taking insulin at regular intervals.

✓ Nowadays insulin can be prepared using techniques of biotechnology.

✓ Insulin was earlier extracted from pancreas of slaughtered cattle and pigs but insulin from these sourcesdevelops allergy or other types of reactions to the foreign protein.

Insulin consists oftwo short polypeptide chains- chain A and chain B, that are linked together by disulphidebridges.

In humans, insulin is synthesised as a pro_hormone,which contains an extra stretch called

C peptide (along with A peptide and B peptide), which is absent in mature insulin.(Figure 12.3) ✓ The Main challenge for production of insulin usingrDNA technique was getting insulin assembled into a mature form.

✓ In 1983 an American company called Eli Lilly first synthesised the genetically engineered insulin.

✓ They prepared two DNA sequences corresponding to A and B polypeptides of insulin.

✓ These DNA Molecules for A and B polypeptides are introduced into separately to E. coli bacteria throughplasmid vectors.

✓ Chains A and B were produced separately, extracted and combined by creating disulphide bondsto form functional human insulin. Gene Therapy v/ It is a collection of methods that allows correction of a gene defect that has been diagnosed in a child or embryo.

✓ The correction of defective gene involves delivery of a normal gene into the individual or embryo to take over the function and compensate for non-functional gene.

✓ The first clinical gene therapy was done in 1990 to a 4 year old girl with adenosine de aminase (ADA) deficiency.

v/ This disorder is caused due to the deletion of the gene for adenosine de-aminase that is essential for immune system to function. Conventional treatment o In some children the disease is cured by bone marrow transplantation.

- o It can be also treated by injecting the functional enzyme (ADA) to the patient. o The problem of above treatments is that they are not completely curative. Gene therapy for ADA Deficiency o As a first step towards gene therapy, lymphocytes from the blood of the patient are grown in a culture outside the body.
- o A functional ADA, cDNA is then introduced into these lymphocytes. o These lymphocytes are then returned into the patient.
- o As these cells (lymphocytes) are not immortal the patient requires periodic infusion of such genetically modified lymphocytes o However, if the gene isolate from bone marrow cells producing ADA is introduced into cells at early embryonic stages, it could be a permanent cure.

Molecular Diagnosis

✓ Conventional method of diagnosis such as serum or urine analysis is not sufficient for the early detectionof disease causing pathogens or virus. ✓ Following methods can be used for early diagnosis. o Recombinant DNA technology o Polymerase Chain Reaction (PCR) o Enzyme Linked Immuno-sorbent Assay (ELISA).

✓ Symptoms of a disease appear only when the concentration of pathogen get increased significantly.

✓ Low concentration of bacteria and virus can be detected by amplification of nucleic acid by PCR.

✓ It detects the mutation in the gene in cancer patients. PCR is routinely used to detect the HI Vin suspected AIDS patients.
Genetic disorder can be also detected by using PCR technique.

✓ A single stranded DNA or RNA, tagged with a radioactive molecule (probe) is allowed to hybridise to its complementary DNA in a clone of cells followed by detection using autoradiography.

✓ The clone having the mutated gene will not appear on the photographic film. ✓ ELISA is based on the principle of antigen-antibody interaction.

✓ Infection by pathogen can bedetected by the presence of antigens like proteins, glycoproteins etc. or by detecting the antibodies synthesised against the pathogen

Transgenic Animals

✓ Transgenic animals are the animals with the modified genome.

v/ A foreign gene is inserted into the genome of the animal to alter its DNA.

✓ Transgenic mice, rats, rabbits, pigs, sheep, cows and fish have been produced. ✓ Common reasons for development of transgenic animals are, Normal physiology and development.

O They are designed to allow the study of gene regulation, their effect on normal functionofthe body.

o By introducing genes from other species that alter the formation of growth factor and studying biological affects that results. Study of a disease.

o A number of transgenic animals are designed to increase our understanding of how genes contribute to the development of disease.

o Transgenic model has been developed for disease like cancer, cystic fibrosis, Alzheimer's disease etc. Biological products o Transgenic animals that produce useful biological products can be created by the introduction of the portion of DNA (gene) which codes for a particular product such as human protein (alpha - I-antitrypsin) used to treat emphysema.

o The first transgenic cow, Rosie, produced human protein-enriched milk (alphalactalbumin). Vaccine safety o Transgenic mice are developed for used in testing the safety of vaccine before they are used on human.

o Polio vaccine was tested on transgenic mice and then on monkey. Chemical safety testing o Transgenic animals are made that carry genes which make them more sensitive to toxicsubstances than non-transgenic animals. o It gives us the results in less time.

Ethical Issues:

v/ The Indian Government has set up organizations such as GEAC (Genetic Engineering Approval Committee), which will make decisions regarding the validity of GM research and the safety ofintroducing GM-organisms for public services. Biopatent:

v/ A patent is the right granted by a government to an inventor to prevent others from making commercial use of his invention.

v/ Now, patents are granted for biological entities and for products derived from biological resources.

Biopiracy:

✓ It is the term used to refer to the use of bio-resources by multinational companies and other organizations without proper authorization from the countries and people concerned,without compensatory payment.

✓ In 1997, an American company got patent rights on Basmati rice through the US Patent and Trademark Office.

This allowed the company to sell a 'new variety of Basmati" in the US and abroad. ✓ This 'new' variety of Basmati had actually been derived from Indian farmer's varieties.

✓ Indian Basmati was crossed with semi-dwarf varieties and claimed as an invention or a novelty.

✓ Several attempts have also been made to patent uses, products and processes based on Indian traditional herbal medicines, Eg. turmeric and neem resources.