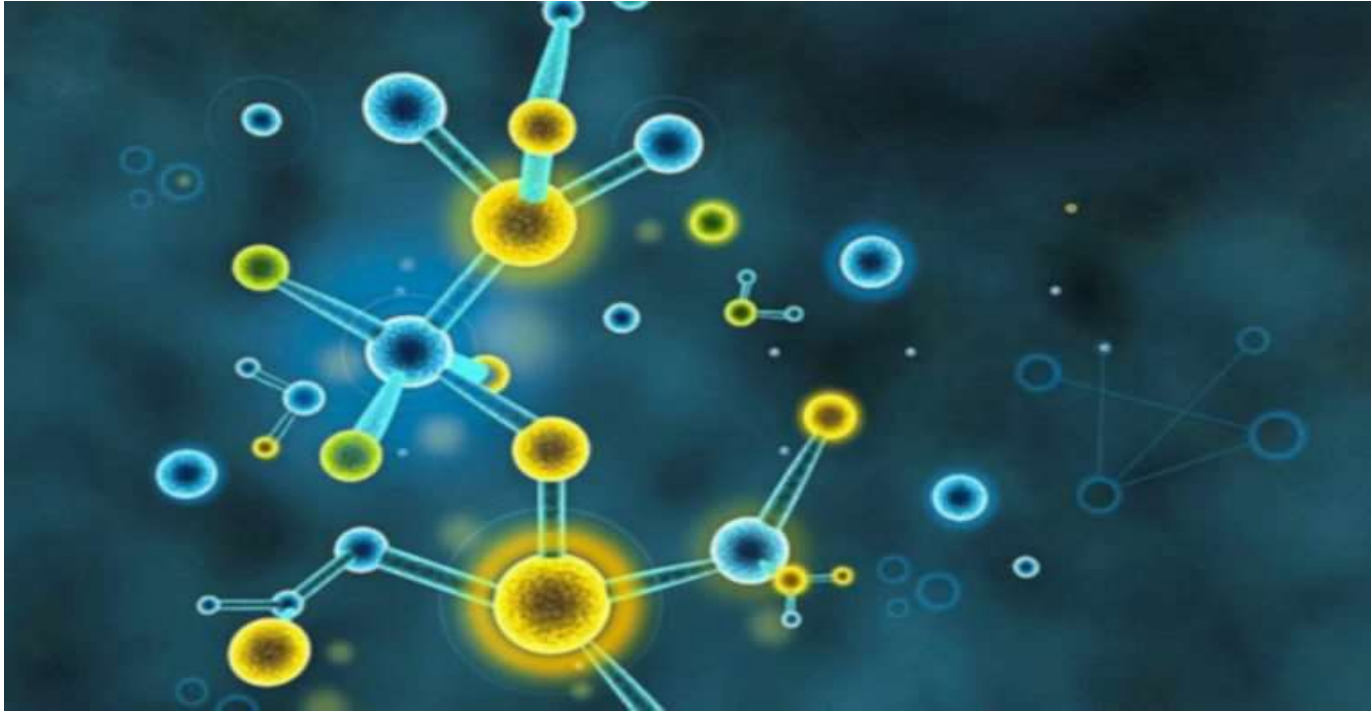




Protein Engineering



Ms. Pratiksha Jayaswal
Assistant Professor
Faculty of Pharmaceutical Sciences,
Rama University, KANPUR (U.P.)

PROTEIN ENGINEERING

- Protein engineering can be defined as the modification of protein structure with recombinant DNA technology or chemical treatment to get a desirable function for better use in medicine, industry and agriculture.

Objectives

- To create a superior enzymes to catalyze the production of high value specific chemicals.
- To produce enzyme in large quantities.
- Eliminate the need for cofactor in enzymatic reaction.
- Change substrate binding sites to increase specificity.
- Change the thermal tolerance and pH stability.
- Increase protein resistance to proteases.
- Rare codon changes.
- To produce biological compounds.
- Investigate how desired mutations can be introduced into a cloned gene.

History



- In 1951 Frederick Sanger discovered how to determine the sequence of amino acids.
- Before this it was thought that proteins have no definite structure. This allowed for translation to be studied as it provided the framework for DNA coding of protein.
- In 1983 Kary Mullis developed the PCR.

RETIONALE OF PROTEIN ENGINEERING

- For industrial application an enzyme, should possess some characteristics in addition to those of enzymes in cells.

These characteristics are :-

(1) enzyme should be robust with long life.

(2) enzyme should be able to use the substrate supplied in the industry even it differs from that in the cell.

(3) enzyme should be able to work under conditions, e.g. extreme of pH, temperature and concentration of the industry even if they differ from those in the cell.

Methods for protein engineering

- A variety of methods are used in protein engineering such as mutagenesis, selection and recombinant DNA technology.

Mutagenesis

- Mutagenesis and selection can be effectively utilized for improving a specific property of an enzyme.
- E.g. for E.coli anthranilate synthetase enzyme is normally sensitive to tryptophan inhibitor due to feedback inhibition but an altered MTR2 mutation of E.coli was found to possess an altered form of enzyme anthranilate synthetase that is insensitive to tryptophan inhibition. And thus helping in the continuous synthesis of tryptophan without inhibition.

Gene Modification

The two processes of gene modification are-

- (a) In vitro mutagenesis using synthetic oligonucleotides.
- (b) Synthesis of complete modified gene de novo.

Chemical modification of enzymes

- The protein synthesized under the control of gene sequence in a cell undergo post-transitional modification. This leads to stability, structural integrity, altered solubility and viscosity of individual proteins.

Protein Engineering Applications

Biotechnological

Biomedical

- ❖ Industrially important enzymes
- ❖ Environmental applications
- ❖ Biomaterials
- ❖ Nanotechnology
- ❖ Biosensors
- ❖ Virus engineering

- ❖ Antibodies
- ❖ Protein Scaffolds
- ❖ Insulin analogs
- ❖ Cardiovascular therapeutics
- ❖ Enzyme based therapeutics
- ❖ Enzyme replacement therapy
- ❖ Cytokines as therapeutics