Subject Code : P8GT11

CORE COURSE XI: GENE EXPRESSION TECHNOLOGY

UNIT- I

Gene expression technology – techniques for gene expression measurement - DNA microarray, RT-PCR, SAGE techniques. Whole genome human DNA microarrays (commercially available microarrays). Whole genome spotted cDNA arrays.

UNIT- II

Heterologous gene expression systems. Expression of heterologous proteins in *E.coli*. Design and construction of expression vectors – recognition and mRNA initiation. Trp, Lac, and tac promoters in direct expression of proteins.- T7 RNA polymerase for direct expression of cloned genes. araB expression systems in *E.coli.*
High level translation initiation – two cistron expression system – vector for enhanced translation of foreign genes in *E.coli*. Gene fusion for expression – trpE fushion. Secretion of heterologous gene products.

UNIT- III

Adenoviral vectors for protein expression – adaptation of adenovirus for gene transfer. Adenoviral expression vectors – construction and applications – advantages and disadvantages.
Expression system in yeast – advantages and disadvantages. *Pitchia pastoris* – Mehylotrophic yeast- AOX1 gene. Molecular genetic manipulation of *P. pastoris*.
Construction of expression vectors – pPICZ. Integration of expression vector into host genome.

UNIT- IV

Recombinant protein expression in plants – promoters – constitution, tissue specific, inducible. Expression of recombinant proteins – pathogens and pest resistance. Bioremediation. Products of recombinant protein expression – oils, fiber, biodegradable plastics, starch, heterologous proteins. Antibody/Antigen production in plants. Virus – mediated expression systems.

UNIT- V

Vectors used for expression in mammalian cells. Assembly of enhancers, promoters, and splice signals to control expression of transferred genes. Cytomegalovirus – promoters for expression in mammalian cells – Siridbis virus – transgenic expression in mice – recombinant expression of proteins in milk of transgenic animals. Selection and coamplification of heterologous genes in mammalian cells. Growth of cell lines – optimizing for production of recombinant proteins.

REFERENCES

1. Abelson, J., D. Goeddel, and M.Simon 1990. Gene Expression Technology. Academic Press, London.
2. Fernandez, J.M.and J.P.Hoeffler. 1999. Gene Expression Systems. Academic Press, London.
3. Causton, H.C., Quackenbush and A. Brazma. 2003. A Beginners Guide: Microarray Gene Expression Data Analysis. Blacwell Publishing, U.K.