#### BIOTECHNOLOGY

## **Unit-I: BASIC TECHNIQUES OF GENETIC MANIPULATION**

Basic techniques: Agarose gel electrophoresis, nucleic acid blotting, transformation of *E. coli*, polymerase chain reaction. Cutting and joining DNA molecules: Restriction enzymes, joining DNA molecules with DNA ligases, adaptors, homopolymer tailing, joining DNA molecules without DNA ligases.

## **Unit-II: PLASMIDS AND PHASE VECTORS**

pBR322, Ti-plasmid Bacteriophage  $\lambda$ , single standard DNA vectors - phage M<sub>13</sub>, cosmids, Phagemids BAC, YAC, Expression vectors, Shuttle vectors.

## Unit-III: CLONING, SCREENING AND SEQUENCING STRATEGIES

Cloning strategies: Cloning genomic DNA - Genomic DNA libraries, PCR as an alternative to genomic cloning. Cloning - Properties cDNA libraries, preparation of cDNA for library construction, full-length cDNA cloning.

## **Unit-IV: SCREENING STRATEGIES**

Sequence - dependent screening, screening by hybridization, probe design, chromosome walking, screening expression libraries - immunological, south-western, north-western blotting, RAPD, RFLP, DNA foot printing.

## **Unit-V: SEQUENCING STRATEGIES**

Basic DNA sequencing - Chain terminator sequencing, automated sequencing, Whole genome sequencing - Analysis of sequence data, DNA sequence databases and data base searches, site-directed mutagenesis. Gene transfer to plants: Agrobacterium mediated transformation, direct DNA transformation - Protoplast transformation, particle bombardment, electroporation and microinjection.

#### References

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# Note: No Practical for this paper.