

Recombinant DNA

Techniques and Tools:

Restriction enzymes

Cloning vectors – extrachromosomal double stranded DNA carried in a host cell.

- Types of vectors
 - plasmids
 - phage
 - cosmids
 - Shuttle vectors
 - BAC – bacteria artificial chromosomes
 - YAC – yeast artificial chromosomes
 - expression vectors
- Must be able to replicate independently – have an origin of replication.
- Must have restriction sites for cloning in fragment DNA.
- Must carry selectable marker(s).
- Must be able to recover vector from the host cell.

Restriction mapping – examples

Ligation

- T4 DNA Ligase
- Alkaline phosphatase

Gel electrophoresis – separation of DNA molecules based on size

PCR – polymerase chain reaction

Library construction

Genomic libraries

cDNA libraries

Library screening