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## Introduction of recombinant DNA into host cells:

Some commonly used procedures:

- 1. Transformation
- 2. Transfection
- 3. Electroporation
- 4. Biolistics
- 5. Agrobacterium mediated gene transfe

DNA is manipulated using various enzymes that modify and/or synthesise it Until 1970 there were no convenient methods available for cutting DNA into discrete, manageable fragments.

1970 - The Beginning of the Revolution Discovery of a restriction enzyme in the bacterium Haemophilus influenzae

Restriction enzymes

• Restriction enzymes are endonucleases

- •Bacterial enzymes.
- •Different bacterial strains express different restriction enzymes.

•The names of restriction enzymes are derived from the name of the bacterial strain they are isolated from.

•Cut (hydrolyse) DNA into defined and **REPRODUCIBLE** fragments.

## •Basic tools of gene cloning .

Names of restriction endonucleases

Titles of restriction enzymes are derived from the first letter of the genus + the first two letters of the species of organism from which they were isolated.

This is known as a Restriction Site The phosphodiester bond is cleaved between specific bases, one on each DNA strand

The product of each reaction is two double stranded DNA fragments Restriction enzymes do not discriminate between DNA from different organisms Restriction endonucleases are a natural part of the bacterial defence system

• Part of the restriction/modification system found in many bacteria

• These enzymes **RESTRICT** the ability of foreign DNA (such as

bacteriophage DNA) to infect/invade the host bacterial cell by cutting it up (degrading it)