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Subject BIOLOGY

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Biotechnology

is a broad area of science involving multiple disciplines designed to use living organisms or their products to perform valuable industrial or manufacturing processes or applications pertaining to human benefit.

Recombinant DNA technology:

An organism's genome contains virtually all the information necessary for its growth and development

Steps in producing recombinant DNA

1. The required gene is cut from a DNA molecule using a restriction enzyme.
2. A bacterial plasmid is isolated and cut with the same restriction enzyme. This ensures cut ends are complementary (same base sequence) to the ends of the required gene.
3. The required gene is joined to the plasmid using the enzyme DNA ligase in a process called ligation.
4. The resulting recombinant plasmid is returned to the bacterial cell.
5. The bacteria reproduce and the required gene is cloned

How do we obtain DNA and how do we manipulate DNA?

Quite straight forward to isolate DNA

For instance, to isolate genomic DNA

1. Remove tissue from organism
2. Homogenize in lysis buffer containing guanidine thiocyanate (denatures proteins)
3. Mix with phenol/chloroform - removes proteins
4. Keep aqueous phase (contains DNA)
5. Add alcohol (ethanol or isopropanol) to precipitate DNA from solution
6. Collect DNA pellet by centrifugation
7. Dry DNA pellet and resuspend in buffer
8. Store at 4°C

Each cell (with a few exceptions) carries a copy of the DNA sequences which make up the organism's genome.

How do we manipulate DNA?

It used to be difficult to isolate enough of a particular DNA sequence to carry out further manipulation and/or characterization of its molecular sequence