Biotechnology Principles and Processes Class 12 Key Points

CBSE Class 12 Biology · NCERT · for boards, NEET and JEE revision

ChapterNotes

What Biotechnology Is

Two core techniques: Genetic engineering + Bioprocess (chemical) engineering

EFB definition covers traditional (curd, bread, wine) and modern (GMOs, recombinant insulin).

- Genetic engineering: alter DNA/RNA and put it in a host to change phenotype
- Bioprocess engineering: keep a sterile, contamination-free setup to grow only the desired microbe at scale
- Inserts ONE chosen gene precisely (breeding mixes thousands of genes)

Five Tools of rDNA Technology

Restriction enzymes · DNA ligase · Cloning vectors · Host · Polymerases (Taq)

Enzymes cutting WITHIN DNA = endonucleases; only at ends = exonucleases.

- Restriction enzymes = molecular scissors; DNA ligase = glue (phosphodiester bonds)
- Vectors carry & replicate foreign DNA; host is usually E. coli
- Foreign DNA must link to an ori → that DNA-with-ori is a replicon

Restriction Endonucleases

EcoRI: 5'–GAATTC–3' / 3'–CTTAAG–5' → cut between G·A → sticky ends

Naming: 1st letter = genus, next two = species (EcoRI = Escherichia coli RY13, I = first isolated).

- Palindrome reads same 5'→3' on both strands
- Sticky-end overhangs base-pair (H-bonds) with any DNA cut by the SAME enzyme
- This is why DNA from two different sources can be joined

Gel Electrophoresis

DNA is (–) charged → moves to anode (+); smaller fragments travel farther

Stained with ethidium bromide → bright orange bands under UV light.

- Medium = agarose; movement depends on fragment size
- Recovering a band cut from the gel = elution
- Used to check cutting and to purify fragments

Cloning Vectors

ori (copy number) + selectable marker (antibiotic resistance) + single recognition site

Plasmids are small, circular, extra-chromosomal DNA that replicate independently.

- ori controls copy number; gene linked to ori is replicated
- Selectable marker eliminates non-transformants, lets transformants grow
- Plants: disarmed Ti plasmid of Agrobacterium tumefaciens; animals: retroviruses

Insertional Inactivation

pBR322: insert at BamHI in tet^R gene → recombinant = tet-sensitive, amp-resistant

Blue-white selection: insert disrupts β-galactosidase → recombinants are WHITE.

- Insertion inactivates the marker, so recombinants lose that resistance
- Selected by replica plating (antibiotic) or chromogenic substrate
- White colony = recombinant; blue colony = non-recombinant

Steps of rDNA Technology

Isolation → Cutting → PCR amplification → Ligation → Insertion (host) → Culture & extract

Isolation enzymes: lysozyme (bacteria), cellulase (plant), chitinase (fungi); chilled ethanol precipitates DNA as fine threads.

- RNase removes RNA, protease removes protein during isolation
- Restriction enzymes cut; ligase joins gene into vector → recombinant DNA
- Recombinant DNA is inserted into a host (transformation), then product is extracted

PCR & Competent Cells

Denaturation (~94°C) → Annealing (2 primers) → Extension (Taq pol) ×~30 → ~10^9 copies

Taq polymerase is thermostable, from Thermus aquaticus.

- Denaturation separates strands; primers flank the target region
- Competent cells made with divalent Ca^{2+} then brief heat shock at 42°C
- Other transfer methods: micro-injection, gene gun, disarmed pathogen vectors

Bioreactors & Downstream Processing

Bioreactor (100-1000+ L): optimum temp · pH · substrate · salts · vitamins · O_2

Stirred-tank type is most common; sparger increases O_2 transfer; foam, temp & pH control plus sampling port.

- Stirred-tank: cylindrical, curved base, agitator mixes & distributes oxygen
- Downstream processing: separation → purification → formulation with preservatives
- Quality control & clinical trials follow; downstream differs product by product

Quick revision

- Restriction enzymes: cut at palindromic sequences → sticky ends; EcoRI (GAATTC)
- Vectors: pBR322 (ori, amp^R, tet^R); Ti plasmid (Agrobacterium, for plants)
- Selectable markers identify recombinants (insertional inactivation, blue-white screening)
- PCR: denature → anneal → extend; Taq polymerase; 2^n copies
- Gel electrophoresis: DNA moves to +ve electrode; smaller = faster
- Gene cloning: isolate → cut → ligate → transform → select → express
- Bioreactors: stirred-tank; downstream processing for purification

Full chapter notes with solved examples and practice questions: https://chapternotes.in/cbse/class-12-biology/biotechnology-principles-and-processes-class-12-notes/