YOUR MENTOR GURU

CSIR NET Life Sciences & Biotechnology — Unit 14

Biotechnology

Study & revision companion | Topic explanations, core facts, and active-recall questions

How to use this guide

Read each topic, explain the key mechanism in your own words, then answer the self-check prompts without looking at the notes. Use your detailed textbook or class notes for full derivations and additional examples.

Topic overview

1. 1. Microscopy and Imaging
2. 2. Centrifugation and Electrophoresis
3. 3. Chromatography and Spectroscopy
4. 4. PCR, Blotting and Sequencing
5. 5. Genomics, Proteomics and Metabolomics
6. 6. Immunological and Binding Assays
7. 7. Experimental Design and Statistics

1. Microscopy and Imaging

Resolution is limited by optics and wavelength; Abbe's relation is approximately d = λ/(2NA) for a simplified case.

Fluorescence microscopy detects labeled structures; confocal microscopy rejects much out-of-focus light.

Electron microscopy uses electron beams for high spatial resolution but requires specialized sample preparation.

Live-cell imaging can reveal dynamics but photobleaching and phototoxicity require control.

Chromatography, electrophoresis and centrifugation separate molecules using different physical properties.

When solving a question, distinguish the underlying mechanism from its observable outcome. Compare the normal situation with the altered condition, and check whether the evidence supports a direct effect or only an association. The central distinction to retain is: sDS-PAGE largely separates denatured proteins by apparent size.

Self-check: Explain the mechanism or relationship above; identify one common misconception; predict the effect of changing a key variable.

2. Centrifugation and Electrophoresis

Differential centrifugation separates components by sedimentation behavior; density gradients improve fractionation.

SDS–PAGE separates denatured proteins primarily by size under common conditions.

Native PAGE retains aspects of native charge and conformation; agarose gels separate nucleic-acid fragments.

RCF depends on rotor radius and rpm: RCF ≈ 1.118×10^{-5} × r(cm) × rpm^2.

UV–visible, fluorescence, NMR, cryo-EM and X-ray methods reveal different molecular properties.

When solving a question, distinguish the underlying mechanism from its observable outcome. Compare the normal situation with the altered condition, and check whether the evidence supports a direct effect or only an association. The central distinction to retain is: beer–Lambert: A = εlc under appropriate assumptions.

Self-check: Explain the mechanism or relationship above; identify one common misconception; predict the effect of changing a key variable.

3. Chromatography and Spectroscopy

Size-exclusion chromatography separates by hydrodynamic size; larger species often elute earlier.

Ion exchange separates molecules according to charge under chosen pH and salt conditions.

Beer–Lambert law A = εlc links absorbance, path length and concentration in its valid range.

Fluorescence and circular dichroism probe different molecular properties; NMR yields structural and dynamic information.

Mass-to-charge measurements support peptide and metabolite identification; tandem MS adds fragmentation evidence.

When solving a question, distinguish the underlying mechanism from its observable outcome. Compare the normal situation with the altered condition, and check whether the evidence supports a direct effect or only an association. The central distinction to retain is: a peak is not automatically a confirmed molecular identity.

Self-check: Explain the mechanism or relationship above; identify one common misconception; predict the effect of changing a key variable.

4. PCR, Blotting and Sequencing

PCR cycles through denaturation, primer annealing and extension; primer design governs specificity.

RT-PCR begins with RNA converted to cDNA; qPCR measures amplification using fluorescence.

Southern blot detects DNA, Northern blot RNA and Western blot protein.

Sanger sequencing uses chain-terminating nucleotides; next-generation platforms sequence many fragments in parallel.

Sequencing characterizes DNA and RNA; single-cell assays resolve cellular heterogeneity.

When solving a question, distinguish the underlying mechanism from its observable outcome. Compare the normal situation with the altered condition, and check whether the evidence supports a direct effect or only an association. The central distinction to retain is: differential expression requires replication and normalization.

Self-check: Explain the mechanism or relationship above; identify one common misconception; predict the effect of changing a key variable.

5. Genomics, Proteomics and Metabolomics

RNA-seq detects transcript abundance and splicing; controls for library size and batch effects are essential.

Mass spectrometry measures mass-to-charge ratios and supports peptide/protein identification.

Targeted metabolomics quantifies selected metabolites; untargeted approaches survey many features.

Single-cell assays reveal cell heterogeneity but can have sparse counts and technical dropout.

Targeted and untargeted methods measure proteins and metabolites; sample preparation affects results.

When solving a question, distinguish the underlying mechanism from its observable outcome. Compare the normal situation with the altered condition, and check whether the evidence supports a direct effect or only an association. The central distinction to retain is: metabolite abundance is not metabolic flux.

Self-check: Explain the mechanism or relationship above; identify one common misconception; predict the effect of changing a key variable.

6. Immunological and Binding Assays

ELISA detects analytes through antigen–antibody binding and an enzyme-linked readout.

Flow cytometry measures light scatter and fluorescence in individual particles or cells.

Surface plasmon resonance can monitor binding kinetics and estimate association and dissociation rates.

Immunoprecipitation enriches a target protein and associated complexes under chosen lysis conditions.

ELISA, western blot, flow cytometry and FISH detect or localize biological molecules.

When solving a question, distinguish the underlying mechanism from its observable outcome. Compare the normal situation with the altered condition, and check whether the evidence supports a direct effect or only an association. The central distinction to retain is: antibody specificity must be checked with controls.

Self-check: Explain the mechanism or relationship above; identify one common misconception; predict the effect of changing a key variable.

7. Experimental Design and Statistics

Randomization reduces systematic allocation bias; blinding can reduce measurement or analysis bias.

A p-value measures compatibility with a null model, not the probability the null hypothesis is true.

Confidence intervals express uncertainty under a specified inferential procedure.

Multiple testing raises false-positive risk; false-discovery-rate procedures help manage it.

Correlation alone cannot establish causation; intervention and controls strengthen inference.

Resolution depends on wavelength and optics; confocal and electron methods address different imaging needs.

When solving a question, distinguish the underlying mechanism from its observable outcome. Compare the normal situation with the altered condition, and check whether the evidence supports a direct effect or only an association. The central distinction to retain is: magnification does not equal resolution.

Self-check: Explain the mechanism or relationship above; identify one common misconception; predict the effect of changing a key variable.

Active-recall practice

1. Explain the essential principles of 1. Microscopy and Imaging and give one experimental or real-world application.

2. Explain the essential principles of 2. Centrifugation and Electrophoresis and give one experimental or real-world application.

3. Explain the essential principles of 3. Chromatography and Spectroscopy and give one experimental or real-world application.

4. Explain the essential principles of 4. PCR, Blotting and Sequencing and give one experimental or real-world application.

5. Explain the essential principles of 5. Genomics, Proteomics and Metabolomics and give one experimental or real-world application.

6. Explain the essential principles of 6. Immunological and Binding Assays and give one experimental or real-world application.

7. Explain the essential principles of 7. Experimental Design and Statistics and give one experimental or real-world application.

Final revision checklist

- 1. Microscopy and Imaging
- 2. Centrifugation and Electrophoresis
- 3. Chromatography and Spectroscopy
- 4. PCR, Blotting and Sequencing
- 5. Genomics, Proteomics and Metabolomics
- 6. Immunological and Binding Assays
- 7. Experimental Design and Statistics

Further study

Use the corresponding unit notes, topic-wise detailed lessons, MCQs, practice tests and full-length mock tests on yourmentorguru.com. This compact PDF is a revision companion, not a replacement for full syllabus textbooks.