

Summary of today's session – Lecture 11 – Protein Techniques & Preamble to Metabolism

Dear Students,

In today's class, we discussed few fundamental concepts related to Techniques to study proteins.

Summary:

Section 1: Re-cap - Overview of Protein Separation Techniques

- Gel filtration columns: composed of porous beads (polyacrylamide, dextran, agarose)
- Gel filtration chromatography: separates proteins based on size (smaller molecules retained longer)
- Separation based on charge differences:
- Proteins interact based on overall charge (positive vs. negative)
- Net charge influenced by varying amounts of positively/negatively charged amino acids and pH
- Separation based on affinity to other molecules:
- Ligand-coupled matrix beads for specific protein interactions
- Protein desorption by excess ligand
- Introduction to protein expression and purification in genetic engineering.

Section 2: Advanced Protein Separation Techniques and Applications

- SDS-PAGE: Separation based on MW
 - Smaller proteins migrate further through gel pores
 - Used for determining subunit composition and MW, comparing with known standards
- Two-dimensional gel electrophoresis (2DE):
 - First dimension: separation based on isoelectric point (pI)
 - Second dimension: separation based on MW using SDS-PAGE
- Mass spectrometry (MS):
 - Protein identification and analysis based on mass-to-charge ratio (m/z)
 - Genome databases aid in identifying gene sequences
- High-throughput (HT) platforms for biomolecular interactions:
 - Microarrays: thousands of discrete proteins for biomarker discovery, protein-protein interactions, and functional characterization
 - Surface plasmon resonance (SPR): measures changes in refractive index for analyzing protein interactions.

Data analysis demonstration

- Short demonstration of R-programming based data analysis

- Application in analyzing and visualizing protein microarray data
- Illustration of computational tools' importance in processing and interpreting biological data
- Emphasis on interdisciplinary nature of modern biological research.

Resource Update:

The course handout and reference materials have been updated and are accessible through the provided Google Drive link:

<https://drive.google.com/drive/folders/1FgzzCom1n6WKlgheQrFLA1U8rkJuISGT>

Our next lecture will discuss metabolism.

Best wishes,
Sanjeeva