Department of Biochemistry

Second Year – Laboratory Technique

Lecture Title: Centrifuge

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Introduction

Centrifugation is a fundamental laboratory technique widely used in biochemistry, molecular biology, and clinical laboratories.
It is based on the application of centrifugal force to separate particles suspended in a liquid medium according to their **density, size, and shape**.

In biochemistry laboratories, centrifugation is essential for sample preparation, purification of biomolecules, and isolation of cellular components. Proper understanding of centrifuge principles and units is critical to ensure accurate and reproducible experimental results.

Principle of Centrifugation

Centrifugation operates on the principle of **sedimentation under centrifugal force**.
When a sample is rotated at high speed, particles experience a force acting outward from the axis of rotation.

- Particles with **higher density or mass** sediment faster and form a **pellet** at the bottom of the tube.
- Lighter components remain suspended in the liquid phase called the **supernatant**.

The sedimentation rate depends on:

- Applied centrifugal force
- Rotor radius
- Particle size and density
- Viscosity of the medium

Types of Centrifuges

1. Clinical Centrifuge

Used in medical and diagnostic laboratories for separation of blood components such as serum and plasma.

2. Bench-Top Laboratory Centrifuge

Commonly used in teaching and research laboratories for routine biochemical assays and sample clarification.

3. Microcentrifuge

Designed for small sample volumes (0.5–2 mL) and frequently used in molecular biology and biochemistry experiments.

4. Refrigerated Centrifuge

Maintains low temperatures during operation to preserve heat-sensitive biomolecules such as enzymes, proteins, and nucleic acids.

5. Ultracentrifuge

Operates at very high speeds and is used for separation of subcellular organelles, viruses, ribosomes, and macromolecules.

Applications of Centrifugation in Biochemistry

- Separation of serum or plasma from blood
- Isolation of cellular organelles (nuclei, mitochondria)
- Protein precipitation and purification
- DNA and RNA extraction
- Removal of cell debris and insoluble materials
- Concentration of biological samples

Units Used in Centrifugation

Two main units are used to express centrifugation conditions:

1. RPM (Revolutions Per Mnute)

RPM represents the speed of rotation of the centrifuge rotor.
It does **not** reflect the actual force applied to the sample and depends only on rotational speed.

2. RCF (Relative Centrifugal Force) or ×g

RCF represents the **true centrifugal force** applied to the sample and is expressed as multiples of gravitational force (g).
It depends on both:

- Rotor speed (RPM)
- Rotor radius

Difference Between RPM and RCF (×g)

Parameter	RPM	RCF (×g)
Measures	Rotational speed	Centrifugal force
Depends on rotor radius	No	Yes
Reflects true force	No	Yes
Preferred in protocols	No	Yes

RCF (×g) is always preferred in biochemical and molecular biology protocols to ensure reproducibility and accuracy.

Relationship Between RPM and RCF

RCF(×g)=1.118×10−5×r×(RPM)2RCF (×g) = 1.118 \times 10^{-5} \times r \times (RPM)^2RCF(×g)=1.118×10−5×r×(RPM)2

Where:

- r = rotor radius (cm)
- RPM = revolutions per minute

Conclusion

Centrifugation is a core laboratory technique in biochemistry that enables efficient separation and purification of biological materials.
Accurate use of centrifuges requires understanding of their types, applications, and correct interpretation of centrifugal units, particularly the distinction between RPM and RCF. Proper centrifugation ensures sample integrity and reliability of experimental data.

References

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