ST. JOSEPH’S COLLEGE FOR WOMEN (AUTONOMOUS) VISAKHAPATNAM

III SEMESTER **BIOCHEMISTRY** TIME: 3Hrs/Week

BCH-Mi1-3801 (3) **ANALYTICAL TECHNIQUES**  Max.Marks:100

w.e.f. 2023-24 admitted batch (23AK) **SYLLABUS**

**Course Objectives: By the end of this course the learner can:**

1. Identify usage of homogenization methods

2. Learn Principles of Electrophoresis and their applications.

3. Gain Knowledge on centrifugation methods and principles involved

4. Understand calorimetry and spectrophotometry

5. Imbibe knowledge on radioisotopes and measurements

**Course Outcomes: On completion of this course students will be able to:**

1. Perform tissue homogenization based on sample sources and use appropriate chromatography techniques

2. Perform electrophoresis and separate samples based on their properties

3. Compare various centrifugation methods available and their principles invovled

4. Illustrate Beer-Lambert’s Law and generate Absorption maxima for biological samples

5. Explain the applications, hazards and precautions of various radioisotopes

**UNIT- I:**

**1. Methods of tissue homogenization and Chromatography**

1.1 Salt and organic solvent extraction and fractionation.

1.2 Dialysis, Reverse dialysis, ultra filtration, lyophilization.

1.3 Chromatography: principle, procedure and application of partition chromatography,

adsorption chromatography, ion exchange chromatography, gel chromatography, affinity

chromatography, GLC and HPLC.

**UNIT – II: 2. Electrophoresis:**

2.1 Basic introduction to free flow, zone electrophoresis.

2.2 Principle, procedure and application of Paper electrophoresis

2.3 Principle, procedure and application of Gel electrophoresis (PAGE, SDS-PAGE and capillary electrophoresis

2.4 Principle, procedure and application of Isoelectric focusing

2.5 Outlines of Principle, procedure and applications of High voltage electrophoresis, Pulse field electrophoresis, Immunoelectrophoresis.

**UNIT – III: 3. Centrifugation**:

3.1 Principle of sedimentation technique.

3.2 Different types of centrifuges and rotors.

3.3 Principle, procedure and application of differential centrifugation,

3.4 Principle, procedure and application of density gradient centrifugation and types,

3.5 Principle, procedure and application of ultra centrifugation,

**UNIT – IV: 4. Colorimetry and spectrophotometry**:

4.1 Laws of light absorption -Beer - Lambert’s law.

4.2 Principle and instrumentation of colorimetry and spectrophotometry.

4.3 Molar extinction coefficient and quantitation.

4.4 Principle of fluorometry and Atomic absorption spectrophotometer

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**UNIT – V:**

**5. Radioisotopes:**

5.1 Important stable radioisotopes used in biochemical research. P32, I 125,I131, Co 60. C 14

etc.

5.2 Radiation hazards and precautions taken while handling radioisotopes.

5.3 Principle and application of RIA.

5.4 Measurement of radioactivity by GM counter

**Reference Books**

1. Physical Biochemistry- Application to Biochemistry and Molecular Biology: Friefelder D. WH Freeman and Company

2. Principles and Techniques of Biochemistry and Molecular Biology: - Ed. K. Wilson and J. Walker, Cambridge University Press.

3. The Tools of Biochemistry: Cooper T.G., John Wiley and Sons Publication.

4. Biophysical chemistry. Principles and Techniques: Upadhayay A, Upadhayay K and Nath N., Himalaya publishing house.

5. Experimental Biochemistry. Cark Jr J. M. and Switzer R.L, W.H. Freeman and Company.

6. Research Methodology for Biological Sciences: Gurumani. N. M.J.P. Publishers., Chennai, India.

7. Instrumental Methods of Chemical Analysis: Chatwal. G and Anand.S., Himalaya Publishing House, Mumbai, India.

7. A Biologist’s Guide to Principles and Techniques of Practical Biochemistry: Williams. B.L. and Wilson. K. (ed.) Edward Arnold Ltd. London

8. Jayaraman, J. (2011). Laboratory Manual in Biochemistry, New Age International (P) Ltd.

9. Sadasivam, S. and Manickam, A. (2005). Biochemical Methods, Second edition, New Age International (P) Ltd.

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