

**SRI VENKATESWARA UNIVERSITY : TIRUPATI**

**MINOR**

**Subject: BIO-CHEMISTRY**

**w.e.f 2024-2025**

**Semester III**

**Course No-2-NALYTICAL TECHNIQUES**

Credits -3

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**COURSE OBJETIVES**

1. To understand the basic concepts of analytical techniques.
2. To gain knowledge about the latest advances in analytical techniques.
3. To apply these techniques in research.

**UNIT-I**

Methods of tissue homogenization. Salt and organic solvent extraction and fractionation. Dialysis, Reverse dialysis, ultra filtration, lyophilization.

Chromatography: principle, procedure and application of partition chromatography, adsorption chromatography, ion exchange chromatography, gel chromatography, affinity chromatography, GLC and HPLC.

**UNIT-II**

Electrophoresis: Principle, procedure and application of free flow, zone electrophoresis (Paper electrophoresis, Gel electrophoresis, PAGE, SDS-PAGE and Disc PAGE). Isoelectric focusing, High voltage electrophoresis, Pulse field electrophoresis, Immunoelectrophoretic.

**UNIT-III**

Centrifugation: Principle of sedimentation technique. Different types of centrifuge and rotors. Principle, procedure and application of differential centrifugation, density gradient centrifugation, ultra centrifugation, rate zonal centrifugation, isopycnic centrifugation.

**UNIT-IV**

Colorimetry and spectrophotometry: Laws of light absorption -Beer - Lambert's law. UV and visible absorption spectra, molar extinction coefficient and quantitation. Principle and instrumentation of colorimetry and spectrophotometry. Principle of nephelometry, fluorometry, Atomic absorption and emission spectrophotometer

**UNIT-V**

Important stable radioisotopes used in biochemical research. P 32, I 125, I 131, Co 60. C 14 etc. Radiation hazards and precautions taken while handling radioisotopes. Principle and application of RIA. Measurement of radioactivity by GM counter and Scintillation counter.

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**Semester III**

**ANALYTICAL TECHNIQUES – (COURSE NO-2)**

Practical

Credits -1

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1. Estimation of ascorbic acid
2. Separation and estimation of total carotenoids and  $\beta$ -carotene
3. Extraction and estimation of vitamin A, vitamin E, niacin and free amino
4. Estimation of phosphorus by Fiske and Subbarow method Characterization of fats– estimation of saponification number, iodine number, acid number and R.M.Number
5. Extraction of Phytoconstituent by Soxhlet and quantification

**COURSE OUTCOMES**

1. After completing this course, the student will
2. Understand the basic concepts and principles of biochemical techniques namely Spectrophotometry, Fluorimetry, Chromatography and Centrifugation.
3. Analyse biochemical compounds such as Carotenoids, Vitamins, Alkaloids and Flavonoids.
4. Identify the compounds by various biochemical techniques and interpret the results
5. Apply the laboratory skills and concepts in carrying out experiments using sophisticated instruments.

**REFERENCE BOOKS**

1. Physical Biochemistry- Application to Biochemistry and Molecular Biology: Friefelder D. WH Freeman and Company 1. Principles and Techniques of Biochemistry and Molecular Biology: - Ed. K. Wilson and J. Walker, Cambridge University Press.
2. The Tools of Biochemistry: Cooper T.G., John Wiley and Sons Publication.
3. Biophysical chemistry. Principles and Techniques: Upadhayay A, Upadhayay K and Nath N., Himalaya publishing house.
4. Experimental Biochemistry. Cark Jr J. M. and Switzer R.L, W.H. Freeman and Company.
5. Research Methodology for Biological Sciences: Gurumani.N. M.J.P. Publishers., Chennai, India.
6. 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.