MODERN PHARMACEUTICAL ANALYSIS

THEORY

GOALS:

The important goals of this course are to give through understanding of the spectroscopy, Mass and chromatographic techniques so that the post graduate students can work in the pharmaceutical companies and research laboratories. Goal of this course is also to train the students structural elucidation of organic compounds.

OBJECTIVES:

On completion of the course, the student shall be able to-

- Know the fundamentals principles, instrumentation and applications of UV-Visible, IR, NMR, Mass spectroscopy, ORD and chromatographic techniques.
- Know ORD, Electrophoresis and statistical analysis.
- Shall be able to analyze drugs and pharmaceuticals using the above instruments.
- Shall be able to interpret the structure of the organic compounds with the given spectral data.
- Shall be able to appreciate the importance of modern instruments in the quality control and research

COURSE CONTENTS

THEORY: 75 hr (3hr/week)

1. UV-VISUAL SPECTROSCOPY: Brief review of electromagnetic spectrum, UV-Visible range, energy, wavelength and color relationships. Interaction of electromagnetic radiation (UV-Visible) with matter and its effects. Chromophores and their interaction with E.M.R. Absorption spectra of organic compounds and complexes illustrating the phenomenon and its utilization in qualitative and quantitative studies of drugs. Shifts and their interpretation (including solvent effects). Empirical correlation of structure with absorption phenomena (Woodward's rules etc), Quantitative estimations, Modern instrumentation. Beer-Lambert Law, Absorption spectra of biomolecules. Theoretical aspects of simultaneous estimation of drugs **6 Hours (10-12 marks)**

2. INFRARED SPECTROSCOPY: Nature of infra-red radiation. Interaction of I.R radiation with oraganic molecules and effects on bonds, Hookes' law. Symmetry rules for observing IR spectrum of molecules. Molecular Infrared spectra. Brief outline of classical I.R instrumentation and particle details of obtaining spectra, including sample preparation for spectroscopy, qualitative interpretation and quantitative applications of FTIR spectra. Theory and applications of FT-IR, ATR and micro ATR. Significance of NIR. **5 Hours (08-10 marks)**

3. OPTICAL ROTATORY DISPERSION: Fundamentals principles of ORD. Cotton effect curves, their characteristics and interpretation. Octant rule and its application with examples. Circular dichroism and its relation to ORD. **3 Hours (05-07 marks)**

4. NUCLEAR MAGNETIC RESONANCE SPECTROSCOPY: Fundamentals principles of NMR (Magnetic properties of nuclei; applied field and precession; absorption and transition; frequency). Chemical shifts concept; isotopic nuclei, Reference standards: Proton Magnetic spectra, their characteristics, presentation terms used in describing spectra and their interpretation (Signal No., Position, Intensity). Brief outline of instrumental arrangements and some practical details. Signal multiplicity phenomenon in high resolution PMR. Spin-spin coupling. Application of Signal Split and coupling constant data to interpretation of spectra. De-coupling and shift reagent methods. Brief outline of principles of FT-NMR with reference to ^{13}C NMR: Spin-spin and spin-lattice relaxation phenomenon. Free induction decay (FID) proton noise de-coupling signal, average time domain and frequency domain signals nuclear overhauser enhancement ^{13}C NMR spectra, their presentation, characteristics, interpretation, examples and applications. Brief indication of application of magnetic resonance spectral data of other nuclei by modern NMR instruments. Introduction to 2-D NMR techniques, TOCSY, NOESY, COSY and INADEQUATE. Methods for Structure determination of small molecules by NMR. **14 Hours (24-27 marks)**

5. MASS SPECTROMETRY: Basic principles and brief outline of instrumentation. Ionization techniques Molecular ion, Meta stable ions, fragmentation processes. Fragmentation patterns and fragmentation characteristics in relation to parent structure and functional groups. Relative abundances of isotopes and their contribution to characteristic peaks. Mass spectrum, its characteristics, presentation and its interpretation. Interfacing a LC and GC in MS **9 Hours (15-17 marks)**

6. CHROMATOGRAPHIC TECHNIQUES: Classification of chromatographic methods based on mechanism of separation. Chromatographic theory, efficiency parameters, Selection of adsorbents and mobile phase for TLC. HPTLC- instrumentation and applications, Progammed multiple development techniques. **4 Hours (07-09 marks)**

7. GAS CHROMATOGRAPHY: Instrumentation, packed and open tubular columns, Liquid stationary phases, derivatization techniques, detectors and critical comparison. Head space GC and temperature programmed GC. Interfacing a GC in FTIR and optical emission spectroscopy. **5 Hours (10-12 marks)**

8. LIQUID CHROMATOGRAPHY: Instrumentation in HPLC, Analytical, preparative and micro-bore columns, ion-exchange columns, size-exclusion columns and chiral columns, Reverse phase HPLC, column selection, mobile phase selection, detectors and its comparison Super fluid critical chromatography and affinity chromatography. Salient features of UPLC. Interfacing a LC in MS & NMR **8 Hours (15-17 marks)**

9. ELECTROPHORESIS: Theory and factors affecting electrophoresis, Moving boundary electrophoresis, zone electrophoresis, Isotachophoresis, Isoelectric focusing and immunoelectrophores, continuous electrophoresis (preparative) application. 2D Gel electrophoresis. **2 Hours (04-06 marks)**

10. X-RAY DIFFRACTION METHODS: Introduction, Generation of X-rays, Elementary crystallography, Miller Indices, X-ray diffraction, Bragg's law, X-ray powder diffraction, X-ray powder diffractometer, obtaining and interpretation of X-ray powder diffraction data and application in polymorphism. **4 Hours (07-09 marks)**

11. THERMO ANALYTICAL METHODS OF ANALYSIS: Thermo gravimetric analysis, Differential Scanning Calorimetry, application in drug interaction and interpretation.

3 Hours (05-07 marks)

12. STATISTICAL ANALYSIS: Introduction, significance of statistical methods, Normal distribution, probability, Degrees of freedom, measures of variation-standard deviation, variance, standard error, tests for statistical significance- students 'T' test, chi-square test. **5 Hours (08-10 marks)**

13. TEACHING SKILLS, RESEARCH METHODOLOGY AND LITERATURE SOURCES: Fundamentals of teaching and learning; art and science of teaching. Thesis writing and presentation of the work. Citation of references. **3 Hours (05-07 marks)**

14. ETHICS IN PHARMACY:

Research Ethics

- Animal and experimental research/humanness
- Human experimentation
- Human volunteer research-informed consent
- Clinical trials
- Gathering all scientific factors
- Gathering all value factors
- Identifying areas of value-conflict, setting priorities
- Working out criteria towards decision
- ICMR/CPCSEA/INSA Guidelines for human / animal experimentation **2 Hours (04-06 marks)**

PRACTICALS: **150 hr (6hr/week)**

Major experiments:

1. Atleast 4 simultaneous estimation of fixed dose combinations by UV-Visible spectroscopic methods.
5. UV-Visible spectrum scanning of certain organic compounds- absorption and correlation of structures. Comparison of UV spectrum of drugs.
6. Comparison of three different analytical methods for three official drugs.
7-8. Experiments based on HPLC and GC. (2 expts)
9. Experimants based on IR
10-15. Structural interpretation of at least 6 different compounds/drugs by UV, IR and Mass data

Minor experiments:

16. Effect of pH and solvent on U.V Spectrum of certain drugs.
17-20. Monograph analysis of paracetamol IP. (minor expts)
21. Separation of amino acids by electrophoresis
22. Any other relevant exercises based on theory.

Text Books

1. Spectrometric Identification of Organic Compounds, 7th Edition Robert M. Silverstein et.al
2. *Fundamentals of Applied Statistics by Gupta* Sc *Kapoor* Vk.
3. Instrumental methods of analysis SKOOG and West and Holler
4. *Willard*, H.H.; *Merrit*, L.L.; *Dean*, J.A.; *Settle*, F.A. *Instrumental Methods of Analysis*,7TH Ed CBS publishers and Distributors, New Delhi,.
5. A.H. Beckett and J.B. Stenlake Practical Pharmaceutical Chemistry, Fourth edition, Part-II
6. Kalsi, P. S. Spectroscopy of Organic compounds, New Age Publishers, New Delhi.
7. http://icmr.nic.in/bioethics/final_cpcsea
8. http://icmr.nic.in/ethical_guidelines

Reference Books

1. *X-Ray methods — Clive* Whiston — *John* Wiley & Sons. 1987
2. Statistics by Gofeti Radhakrishna
3. Biostastistics by Sadkar
4. Instrumental methods of analysis Scoog and West
5. Instrumental Method of Analysis - modern methods Part-B, Vol-2, pages 11to 54 Editor by J.W.*Munson* (Marcel Dekker) 4.
6. Kemp William, Organic spectroscopy, Pal Grave, New York.
7. Kalsi, P. S. Spectroscopy of Organic compounds, New Age Publishers, New Delhi.
8. Willard HH, Merritt LL Dean, JA, Settle FA: Instrumental methods of. Analysis, 7th edition, CBS publishers and Distributors, New Delhi,.
9. Robert M. Silverstein, Spectrometric Identification of Organic Compounds, Willey
10. Stahl, E.*:* Thin-Layer Chromatography, A Laboratory Handbook, 2nd. Edit.,
11. Modern NMR Techniques for Chemistry Research. A. E. Derome. Pergamon Press

Journals:

At least one International journal is to be subscribed

Teaching/ Learning activities:

1. Journal Club: Minimum of one presentation per term per student and evaluation is compulsory.
2. Seminar: Minimum of one seminar per term per student.
3. Field visits/ Industrial visits: Minimum of one visit during first year.
4. Conferenc/ meeting in their respective discipline.

Scheme of the examination:

Subjects	**Sessionals**	**Seminar/record marks**	**Annual Examination Marks**	**Total marks**
Theory	**30**	**20 (Journal Club/ Seminar)**	**100 (3 Hours)**	**150**
Practical Record	**30**	**20 (Seminar/record marks)**	**100 (6 Hours)**	**150**

University practical Examination

Synopsis	10 marks
Major Experiment	40 marks
Minor Experiment	30 marks
Viva-Voce	20 marks
Total	100 marks