SCHEME : J

Name : _____

Roll No. : _____ Year : 20___ 20___

Exam Seat No. : _____

LABORATORY MANUAL FOR
PHARMACEUTICAL CHEMISTRY
(20052)

FIRST YEAR D.PHARMACY

MAHARASHTRA STATE BOARD OF TECHNICAL EDUCATION, MUMBAI
(Autonomous) (ISO 9001: 2015) (ISO/IEC 27001:2013)

VISION

To ensure that the diploma level technical education constantly matches the latest requirements of technology and industry and includes the all-round personal development of students including social concerns and to become globally competitive, technology led organization.

MISSION

To provide high quality technical and managerial manpower, information and consultancy services to the industry and community to enable the industry and community to face the challenging technological & environmental challenges.

QUALITY POLICY

We, at MSBTE are committed to offer the best-in-class academic services to the students and institutes to enhance the delight of industry and society. This will be achieved through continual improvement in management practices adopted in the process of curriculum design, development, implementation, evaluation and monitoring system along with adequate faculty development programmes.

CORE VALUES

MSBTE believes in the following:

- ✓ Skill development in line with industry requirements.
- ✓ Industry readiness and improved employability of Diploma holders.
- ✓ Synergistic relationship with industry.
- ✓ Collective and Cooperative development of all stake holders.
- ✓ Technological interventions in societal development.
- ✓ Access to uniform quality technical education.

LABORATORY MANUAL OF

PHARMACEUTICAL CHEMISTRY

(20052)

First Year

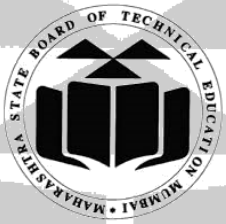
Diploma in Pharmacy

Maharashtra State Board of Technical Education, Mumbai.

(Autonomous)

(ISO 9001:2015) (ISO/IEC27001:2013)

PCI ER-2020/'J' Scheme Curriculum



Maharashtra State Board of Technical Education, Mumbai

(Autonomous) (ISO 9001:2015) (ISO/IEC27001:2013)

4th floor, Government Polytechnic Building, 49, Kherwadi, Bandra (E), Mumbai- 400 051

(Printed on – July 2024)

MAHARASHTRA STATE BOARD OF TECHNICAL EDUCATION, MUMBAI

CERTIFICATE

This is to certify that Mr. /Ms. _____

Roll No. _____ of First Year Diploma in Pharmacy studying at _____

has completed the practical work satisfactorily in Pharmaceutical Chemistry (20052) for the academic year 20 - 20 as prescribed in the PCI ER 2020 syllabus.

Date: _____ Enrollment No.: _____

Place: _____ Exam Seat No.: _____

Course Teacher **Principal**

External Examiner

Seal of the Institute

MAHARASHTRA STATE BOARD OF TECHNICAL EDUCATION MUMBAI

PROGRAM OUTCOMES

1. **Pharmacy knowledge:** Possess knowledge and comprehension of the core and basic knowledge associated with the profession of pharmacy.
2. **Modern tool usage:** Learn, select, and apply appropriate methods and procedures, resources, and modern pharmacy-related computing tools with an understanding of the limitations.
3. **Leadership skills:** Understand and consider the human reaction to change, motivation issues, leadership and team-building when planning changes required for fulfilment of practice, professional and societal responsibilities. Assume participatory roles as responsible citizens or leadership roles when appropriate to facilitate improvement in health and wellbeing.
4. **Professional identity:** Understand, analyze and communicate the value of their professional roles in society (e.g. health care professionals, promoters of health, educators, managers, employers, employees).
5. **Pharmaceutical ethics:** Honour personal values and apply ethical principles in professional and social contexts. Demonstrate behavior that recognizes cultural and personal variability in values, communication and lifestyles. Use ethical frameworks; apply ethical principles while making decisions and take responsibility for the outcomes associated with the decisions.
6. **Communication:** Communicate effectively with the pharmacy community and with society at large, such as, being able to comprehend and write effective reports, make effective presentations and documentation, and give and receive clear instructions.
7. **The Pharmacist and society:** Apply reasoning informed by the contextual knowledge to assess societal, health, safety and legal issues and the consequent responsibilities relevant to the professional pharmacy practice.
8. **Environment and sustainability:** Understand the impact of the professional pharmacy solutions in societal and environmental contexts, and demonstrate the knowledge of, and need for sustainable development.
9. **Life-long learning:** Recognize the need for and have the preparation and ability to engage in independent and life-long learning in the broadest context of technological change. Self-assess and use feedback effectively from others to identify learning needs and to satisfy these needs on an ongoing basis

COMPETENCIES FOR THE INDIAN D. PHARM HOLDERS

Competency is defined as "A distinct composite of knowledge, skill, attitude and value that is essential to the practice of the profession in real life contexts".

The candidates who successfully complete the Diploma in Pharmacy (D. Pharm) program of Education Regulations 2020 (ER-2020), from the institutions approved by the Pharmacy Council of India are expected to attain the following professional competencies.

1. Review Prescriptions: The student should receive and handle prescriptions in a professional manner and be able to check for their completeness and correctness. Also, the prescribers should be contacted for any clarifications & corrections in the prescriptions with suggestions if any.

2. Dispense Prescription / Non-Prescription Medicines: The student should be able to dispense the various scheduled drugs / medicines as per the implications of the Drug & Cosmetic Act and Rules thereunder. Also, the non-prescription medicines (over-the-counter drugs) should be dispensed judicially to the patients as required.

3. Provide Patient Counselling / Education: The student should be able to effectively counsel / educate the patients / caretakers about the prescription / non-prescription medicines and other health related issues. Effective communication includes using both oral and written communication skills and various communication techniques.

4. Hospital and Community Pharmacy Management: The student be able to manage the drug distribution system as per the policies and guidelines of the hospital pharmacy, good community pharmacy practice and the recommendations of regulatory agencies. Also, be able to manage the procurement, inventory, and distribution of medicines in hospital / community pharmacy settings.

5. Expertise on Medications: The student should be able to provide an expert opinion on medications to health care professionals on safe and effective medication – use, relevant policies and procedures based on available evidence.

6. Proficiency on Pharmaceutical Formulations: The student should be able to describe the chemistry, characteristics, types, merits and demerits of both drugs and excipients used in pharmaceutical formulations based on her/his knowledge and scientific resources.

7. Entrepreneurship and Leadership: The student should be able to acquire the entrepreneurial skills in the dynamic professional environments. Also, be able to achieve leadership skills through teamwork and sound decision-making skills.

8. Deliver Primary and Preventive Healthcare: The student should be able to contribute to various healthcare programs of the nation including disease prevention initiatives to improve public health. Also contribute to the promotion of national health policies.

9. Professional, Ethical and Legal Practice: The student should be able to deliver professional services in accordance with legal, ethical, and professional guidelines with integrity.

10. Continuing Professional Development: The student should be able to recognize the gaps in the knowledge and skills in the effective delivery of professional services from time to time and be self-motivated to bridge such gaps by attending continuing professional development programs.

COMPETENCY MAPPING WITH THE COURSE

Competencies	Pharmaceutical Chemistry
1.Review Prescriptions	✓
2. Dispense Prescription / Non-Prescription Medicines	✓
3.Provide Patient Counselling / Education	✓
4.Hospital and Community Pharmacy Management	
5.Expertise on Medications	✓
6.Proficiency on Pharmaceutical Formulations	✓
7.Entrepreneurship and Leadership	
8.Deliver Primary and Preventive Healthcare	
9.Professional, Ethical and Legal Practice	
10.Continuing Professional Development	✓

GRAPHICAL STRUCTURE OF SUBJECT AREA

PHARMACEUTICAL CHEMISTRY (20052)

APPLICATION / PROBLEM

Students will be able to:

- Identify organic compound, cation and anion present in given sample.
- Synthesize organic compound or derivative.
- Analyse the given drug sample and determine its limits of purity as per Indian Pharmacopoeia.
- Prepare standard solutions using the principles of volumetric analysis.
- Perform volumetric analysis.

↑

PROCEDURE

Systematic qualitative analysis.
Identification tests / Limit tests as per I. P.
Synthesis of derivatives or organic compounds.
Volumetric analysis and different titration methods.

↑

PRINCIPLE

Basic chemical reactions like oxidation, reduction, esterification, hydrolysis, bromination, acetylation, nitration etc.
Principles of Volumetric analysis and different titration methods.

↑

CONCEPT

Solubility, Colour, Odour, Acidity & Basicity, Functional groups, Volumetry, Synthesis.

↑

FACTS

Qualitative & Quantitative analysis of Organic compounds, Cations and Anions. Impurity testing.

PHARMACEUTICAL CHEMISTRY – PRACTICAL

Course Code: ER20-12P/20052 **75 Hours (3 Hours / Week)**

Scope: This course is designed to impart basic training and hands-on experiences to synthesis chemical substances used as drugs and pharmaceuticals. Also, to perform the quality control tests, impurity testing, test for purity and systematic qualitative analysis of chemical substances used as drugs and pharmaceuticals.

Course Objectives: This course will provide the hands-on experience on the following aspects of chemical substances used as drugs and pharmaceuticals

1. Limit tests and assays of selected chemical substances as per the monograph
2. Volumetric analysis of the chemical substances
3. Basics of preparatory chemistry and their analysis
4. Systematic qualitative analysis for the identification of the chemical drugs

Course Outcomes: Upon successful completion of this course, the students will be able to

1. Perform the limit tests for various inorganic elements and report
2. Prepare standard solutions using the principles of volumetric analysis
3. Test the purity of the selected inorganic and organic compounds against the monograph standards
4. Synthesize the selected chemical substances as per the standard synthetic scheme
5. Perform qualitative tests to systematically identify the unknown chemical substances

Practicals

Sr. No.	Experiment
1	**Limit test for** • Chlorides; sulphate; Iron; heavy metals
2	Identification tests for Anions and Cations as per Indian Pharmacopoeia
3	**Fundamentals of Volumetric analysis** Preparation of standard solution and standardization of Sodium Hydroxide, Potassium Permanganate
4	**Assay of the following compounds** • Ferrous sulphate-by redox titration • Calcium gluconate-by complexometric • Sodium chloride-by Modified Volhard's method • Ascorbic acid by iodometry • Ibuprofen by alkalimetry
5	**Fundamentals of preparative organic chemistry** Determination of Melting point and boiling point of organic compounds
6	**Preparation of organic compounds** • Benzoic acid from Benzamide • Picric acid from Phenol
7	**Identification and test for purity of pharmaceuticals** Aspirin, Caffeine, Paracetamol, Sulfanilamide
8	Systematic Qualitative analysis experiments (4 substances)

ASSIGNMENTS

The students shall be asked to submit the written assignments on the following topics (One assignment per student per sessional period i.e., a minimum of THREE assignment per student)

1. Different monographs and formularies available and their major contents
2. Significance of quality control and quality assurance in pharmaceutical industries
3. Overview on Green Chemistry
4. Various software programs available for computer aided drug discovery
5. Various instrumentations used for characterization and quantification of drug

STRATEGY FOR IMPLEMENTATION

It is suggested that 32-35% experiments shall be completed before every sessional exam.

GUIDELINES FOR TEACHERS

Teacher shall explain the following points to the students before starting of the practical:

1. **Learning Objectives:** To foster better understanding of the subject and to inculcate the skills and attitude related practicals.
2. **Graphical structure:** In graphical structure topics and subtopics are organized in systematic way so that ultimate purpose of learning the subject is achieved. This is arranged in the form of fact, concept, principle, procedure, application and problem.
3. **Elementary Guide to work in Laboratory:** The methods and other finer details of the equipment including equipment specifications should be explained to avoid equipment breakages, create conducive environment for proper organizing of the practical work with the time schedule.
4. Teachers should verify and check the work conditions of the equipment and request the students to follow the standard operating procedures (SOP).
5. Before starting the practical, Teachers should explain the strategies of the experiment.
6. Teachers should ensure the active participation of students while performing the experiment.
7. Observations should be checked individually, and each student should be given a chance to perform the experiment.
8. Teachers should ask the students to complete the questions which are given at the end of the experiment accordingly.
9. Assessment of manuals should be done according to the assessment norms. Proper marks should be distributed according to the performance of the individuals.
10. Teachers should explain the competencies that student should achieve, in detail with their importance to students after completion of their course.
11. Apart from the syllabus, teachers should provide and cover extra topics which are beneficial for the students.
12. Explanation about various equipment with some interesting videos, reagents, chemicals, glassware should be given to students prior to commencing of the practical.
13. Teachers should observe the students when students are performing practicals in groups, proper contributions of the individual student should be there, and record of observation should be noted by all of them.
14. Teachers should also organize a visit to the pharmaceutical industries where students get a brief idea about the manufacturing processes of common dosage forms such as tablets, capsules, liquid orals, injectables, etc.
15. Teachers should also ask them to gather information about each type of dosage forms, their generic name, branded names and label contents.
16. Teachers may suggest the students to refer to sources of information such as literature, research papers, books, attending conferences, seminars for the updating of knowledge.
17. According to the professional competencies given by PCI, teachers should develop the professional skills of the students.
18. Teacher should construct different types of sessions for students such as quiz, group discussions projects on different topics, etc.
19. Teachers should ensure that revised CIAAN – 2017 norms or the latest norms given by MSBTE are followed simultaneously and implemented.
20. Teachers should follow the guidelines given by PCI & MSBTE from time to time.

BLOOMS TAXONOMY LEVELS

1 Knowledge

Define, Identify, Describe, Recognize, Tell, Explain, Recite, Memorize, Illustrate, Quote

2 Understand

Summarize, Interpret, Classify, Compare, Contrast, Infer, Relate, Extract, Paraphrase, Cite

3 Apply

Solve, Change, Relate, Complete, Use, Sketch, Teach, Articulate, Discover, Transfer

4 Analyze

Contrast, Relate, Devise, Distill, Correlate, Illustrate, Conclude, Categorize, Connect, Take apart

5 Evaluate

Criticize, Reframe, Judge, Defend, Appraise, Value, Prioritize, Plan, Grade,

6 Create

Design, Modify, Role-play, Develop, Rewrite, Pivot, Modify, Collaborate, Invent, Write

INSTRUCTIONS FOR STUDENTS

Students should follow the instructions given below for better understanding of the subject from a theoretical and practical concept of view.

1. As per the instructions, the students should wear an apron, cap, mask, gloves and slippers before entering the lab.
2. The students should keep their important things in the locker which is provided by the college.
3. While entering the laboratory, the students should carry manual, rough book and practical requirements as instructed.
4. Students should attend the practical regularly throughout the year, so as to understand the subject properly, and to develop the skills for performing the experiments and attaining the competencies.
5. The students should carry out the experiment individually and perform the experiment at the allotted specific work area.
6. The practical applications of every experiment should be noted by the students.
7. Students should answer the questions asked in the practicals and should ask the teacher about their difficulties without any hesitation.
8. After completion of practicals students should write the answers of the question given at the end of the experiment.
9. Students should develop different types of competencies to become competent Pharmacists.
10. Students should actively participate in group discussions, activities, etc. and strive to achieve the knowledge, skills, and attitude.
11. Student should submit the manual for assessing regularly on the scheduled date.
12. After completing the practical, the student should clean the platform and glassware that he has used.

LABORATORY MANUAL OF PHARMACEUTICAL CHEMISTRY

MAPPING OF COURSE OUTCOMES

Expt No.	Title of Experiment	CO1	CO2	CO3	CO4	CO5
01	Introduction to laboratory & study of laboratory chemicals, equipments & glasswares	✓	✓	✓	✓	✓
02	Limit test for Chloride	✓	✓	✓		
03	Limit test for Sulphate	✓	✓	✓		
04	Limit test for Iron	✓	✓	✓		
05	Limit test for Heavy metals	✓	✓	✓		
06	Identification tests for Anions			✓		✓
07	Identification tests for Cations			✓		✓
08	Preparation and standardization of Sodium hydroxide solution		✓	✓		
09	Preparation and standardization of Potassium permanganate solution		✓	✓		
10	Assay of Ferrous sulphate		✓	✓		
11	Assay of Calcium gluconate		✓	✓		
12	Assay of Sodium chloride		✓	✓		
13	Assay of Ascorbic acid		✓	✓		
14	Assay of Ibuprofen		✓	✓		
15	Determination of Melting point			✓		
16	Determination of Boiling point			✓		
17	Synthesis of benzoic acid from benzamide				✓	
18	Synthesis of Picric acid from phenol				✓	
19	Identification test of Aspirin					✓
20	Identification test of Caffeine					✓
21	Identification test of Paracetamol					✓
22	Identification test of Sulphanilamide					✓
23	Systematic Qualitative Analysis- Compound 1					✓
24	Systematic Qualitative Analysis- Compound 2					✓
25	Systematic Qualitative Analysis- Compound 3					✓
26	Systematic Qualitative Analysis- Compound 4					✓

LIST OF EXPERIMENTS AND RECORD OF PROGRESSIVE ASSESSMENT

Expt No.	Title of Experiment	Page No.	Date of Performance	Date of Submission	Assessment Marks	Sign of Teacher
01	Introduction to laboratory	1				
02	Limit test for Chloride	9				
03	Limit test for Sulphate	13				
04	Limit test for Iron	17				
05	Limit test for Heavy metals	21				
06	Identification tests for Anions	25				
07	Identification tests for Cations	33				
08	Preparation and standardization of Sodium hydroxide solution	42				
09	Preparation and standardization of Potassium permanganate solution	46				
10	Assay of Ferrous sulphate	51				
11	Assay of Calcium gluconate	56				
12	Assay of Sodium chloride	62				
13	Assay of Ascorbic acid	68				
14	Assay of Ibuprofen	74				
15	Determination of Melting point	79				
16	Determination of Boiling point	83				
17	Synthesis of benzoic acid from benzamide	87				
18	Synthesis of Picric acid from phenol	92				
19	Identification test of Aspirin	97				
20	Identification test of Caffeine	102				
21	Identification test of Paracetamol	106				
22	Identification test of Sulphanilamide	110				
23	Systematic Qualitative Analysis- Compound 1	133				
24	Systematic Qualitative Analysis- Compound 2	140				
25	Systematic Qualitative Analysis- Compound 3	147				
26	Systematic Qualitative Analysis- Compound 4	154				

I) PRACTICAL RECORD MARKS*:

Sessional Exam	Experiment No.		Total no. of experiment conducted	Average marks obtained for the experiment conducted. (out of 10)	Teacher's Signature
	From	To			
First Sessional					
Second Sessional					
Third Sessional					

*Sessional wise marks should be considered for internal assessment of practical sessional examinations (out of 10M)

II) ASSIGNMENT MARKS#:

Sr. No.	Title of Assignment	Marks out of 10#	Assignment Marks (Average of three)	Teacher's Signature
1				
2				
3				

#Marks should be transferred from Appendix -1 A typical format for assessment of an assignment.

Average Sessional Mark out of 10	Assignments Mark out of 10 (Average of three)	Total Marks out 20	Teacher's Signature

Experiment No. 1
Introduction to laboratory-Study of laboratory Chemicals, Glasswares & Equipment

1. Aim
To prepare a table of laboratory chemicals, glass wares and equipment along with uses and precautions.

2. Practical Significance
The appropriate uses of chemicals, glassware, and equipment ensure experiments are conducted correctly. The potential hazards, proper handling techniques, and safety protocols relevant to their laboratory work.

3. Practical Outcomes (PrO)
After completion of this practical, the students will be able to:

PrO	Practical Outcomes	Mapped CO	BTL
1	Record the uses and hazards of laboratory chemicals.	CO 1-5	3
2	Handle various equipment used in the laboratory.	CO 1-5	2
3	Follow safety rules	CO 1-5	3
4	Follow precautions in handling chemicals.	CO 1-5	3

4. Relevant Theoretical Background
A. Chemicals
Chemicals are substances with defined chemical compositions that undergo chemical reactions. Understanding chemical properties, such as reactivity, solubility, and toxicity, is crucial for safe handling and proper use.

B. Glassware
Glassware is commonly used in laboratories for holding, measuring, mixing, and observing substances. Different types of glassware include beakers, flasks, test tubes, and pipettes, each with specific functions and volume measurements.

The pharmaceutical chemistry laboratory requires different glassware for smooth conduction of practical such as Burette, Pipettes (graduated and Volumetric), Conical flask, Measuring cylinder, Beaker, Volumetric flask, Test tube, Reagent bottle, Nessler's cylinder, Funnel, Glass rod, Reflux condenser, round bottom flask etc.

C. Equipment
a. Balances
Uses: Precisely measure the mass of substances for experiments and formulations.

Precautions: Calibrate balances regularly, handle with care to avoid damage to sensitive components, and use anti-static measures to prevent interference with measurements.

There are various balances with varied degrees of sensitivity are as follows:

Physical balance, Analytical balance, Triple beam balance, Torsion type balance, Single pan electronic balance, Platform electronic balance.

b. Autoclaves
Uses: Sterilize laboratory equipment, media, and waste by subjecting them to high-pressure steam.

Precautions: Follow loading instructions to ensure proper steam circulation, monitor pressure and temperature during operation, and allow adequate cooling time before opening to avoid burns.

c. pH Meters

Uses: Measure the acidity or alkalinity of a solution by detecting hydrogen ion concentration.
Precautions: Calibrate pH meters using standard buffer solutions, handle electrodes with care to avoid damage or contamination, and clean and store properly after each use.

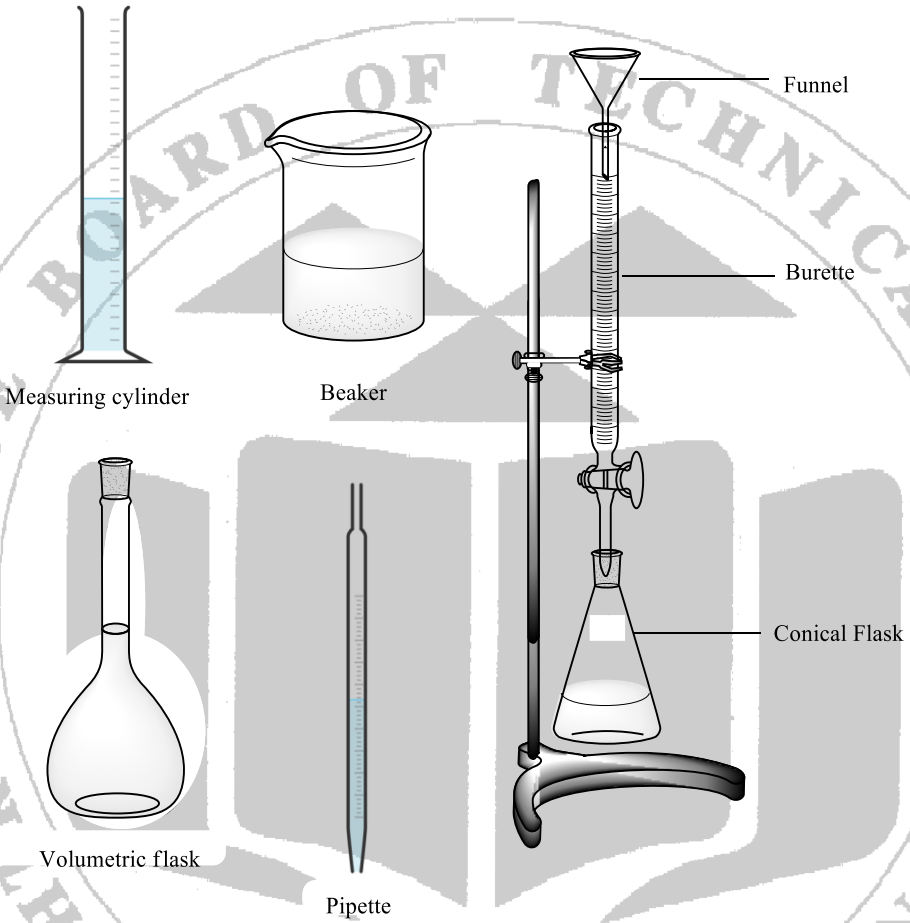


Fig1.1: Common laboratory apparatus

d. Microscopes

Uses: Magnify and visualize small objects or structures for observation and analysis.
Precautions: Handle slides and coverslips carefully to avoid breakage and contamination, use appropriate magnification and lighting settings for the sample being observed, and clean lenses regularly to maintain clarity.

e. Incubators

Uses: Provide controlled temperature and humidity conditions for growing cultures or conducting experiments requiring specific environmental conditions.
Precautions: Monitor and calibrate temperature and humidity settings regularly, avoid overcrowding incubators to ensure proper air circulation, and clean and disinfect interior surfaces between uses.

f. Hot Air Oven

Uses: Utilized for sterilizing and drying laboratory equipment, glassware, and heat-resistant materials by exposing them to dry heat at controlled temperatures.

Precautions: Ensure the oven is set to the appropriate temperature and use a calibrated thermometer to verify accuracy regularly. Arrange items inside the oven to promote even heating and airflow, avoiding overcrowding.

g. Vacuum Filter

Uses: In chemical synthesis or pharmaceutical manufacturing, vacuum filtration is used to isolate products from reaction mixtures or purification processes at controlled pressure.

Precautions: Clean the filtration apparatus thoroughly after each use to prevent contamination and ensure optimal performance. Ensure there are no air bubbles trapped in the filter paper or filter funnel, as they can reduce filtration efficiency and cause uneven filtering.

h. Fuming cupboard

Uses: Fuming cupboards, also known as fume hoods or fume cabinets, are primarily used for handling hazardous chemicals that emit toxic fumes, vapours, or gases. They provide a controlled environment to safely contain and vent these hazardous substances.

Precautions: Ensure the fuming cupboard is properly connected to an exhaust system that effectively removes hazardous fumes. Wear appropriate PPE, including lab coats, gloves, and safety goggles, when working inside the fume hood to protect against chemical exposure.

These are a few examples of laboratory equipment and some associated precautions. Proper training, maintenance, and adherence to safety protocols are essential for ensuring the accurate and safe operation of laboratory equipment.

5. Requirements

a. Common glasswares and chemicals.
b. Common laboratory equipment.

6. Requirements Used

_____

_____

7. Procedure

a. Listen carefully to the lecture given by teacher about the practical significance of subject, relevant professional competencies, practical learning outcomes, and skills to be developed, information about chemicals, glassware and equipment, method of continuous assessment and tentative plan of work in the laboratory.
b. Accompany the teacher on a tour of the laboratory, where you'll visit the balance room and fume cupboard, gaining insight into the overall laboratory operations.
c. Observe the equipment and record the information in the observation table.
d. Observe the charts and diagrams displayed in the laboratory.
e. Understand the general precautions to be followed while working in the laboratory.
f. Seek out demonstration of fire extinguisher mounted on the wall of the laboratory from the teacher.

8. Observations

Table 1: Common Chemicals

Sr. No.	Name and formula	Use	Remarks/Hazards/Precautions if any
1.			
2.			
3.			
4.			
5.			

Table 2: Common Reagents

Sr. No.	Name of reagents	Use	Remarks/Hazards/Precautions if any
1.			
2.			
3.			
4.			
5.			

Table 3: Common Glasswares

Sr. No.	Name of glasswares	Use	Remarks/Hazards/Precautions if any
1.			
2.			
3.			
4.			
5.			

Table 4: Common Equipment

Sr. No.	Name of equipment	Use	Remarks/Hazards/Precautions if any
1.			
2.			
3.			
4.			
5.			

9. References

a. Indian Pharmacopoeia 2022.

10. Practical Related Questions

a. Write a note on fire hazards in a pharmaceutical chemistry laboratory.
b. Define Laboratory grade reagent and Analytical grade reagent.
c. Enlist five essential requirements you must have for performing practical in the chemistry laboratory.
d. What is the use of a fume cupboard in the chemistry laboratory?
e. List inflammable chemicals in the laboratory.

(*Space for Answers*)

MAHARASHTRA STATE BOARD OF TECHNICAL EDUCATION MUMBAI

11. Assessment Scheme

Particular	Understanding the basic concept (Intellectual skill)	Activities (Motor skill)	Cleanliness & Handling (Affective domain)	Viva-voce / Answers Written	Total	Signature of teacher
Marks Obtained						
Max Marks	**02**	**05**	**01**	**02**	**10**	

LIMIT TEST

Limit = It is a value or quantity that is present in the compound/ drug.
Test = Simply means to investigate or examine.

The drugs or active pharmaceutical ingredient (API) intended for human use should be pure, safe, and, efficacious. Numerous sources of impurities arise in the chemical compounds. The impure drugs compromise the safety and efficacy of the formulations.

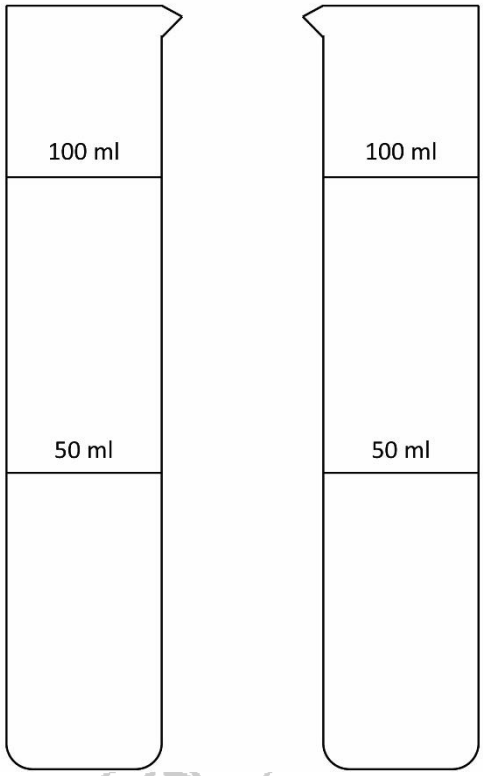
A limit test is defined as a quantitative or semi-quantitative test designed to identify and control a small quantity of impurity that is likely to be present in the substance.

A limit test is generally implied to find out the inorganic impurities that are present in the substance. Official books of standards such as Indian Pharmacopoeia (IP), have fixed permissive limits of tolerance for impurities in different drugs. The drug/ compound shall comply with the limits mentioned in the standard books such as IP.

Sources of impurities in pharmaceuticals may arise from

1) Raw material itself
2) Defects in the manufacturing process of the pharmaceutical compounds
3) Chemical decomposition/ degradation in the presence of moisture or light.
4) Deliberate adulteration
5) Careless storage
6) Improper packing

In this section, we are going to perform the limit test for the chlorides, sulphates, iron, and heavy metals of various inorganic and organic pharmaceuticals. Presence of the chlorides, sulphates, iron, heavy metals and other impurities beyond certain limits have undesirable/ adverse effects on the human body.



Limit tests are conducted using Nesssler's cylinders, which are crafted from colorless glass, adhering to precise dimensions outlined in Pharmacopoeias. Typically, these cylinders boast a 50 mL capacity and stand at a height of 150 mm. The 50 mL demarcation is clearly indicated on the cylinder's neck. To ensure consistency, two identical cylinders are utilized—one for the 'Test' (sample) and the other for the 'Standard,' facilitating direct comparison. Distilled water serves as the sole medium for conducting limit tests, as tap water contains numerous ions that could interfere with the accuracy of the results.

Fig: Pair of Nessler's Cylinders

Experiment No. 2
Limit Test for Chloride

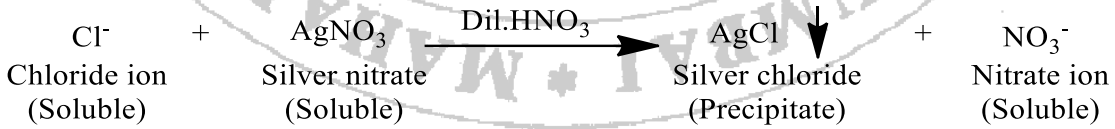
1. Aim
To perform and report the limit test for chloride on the given samples as per IP-2022.

2. Practical Significance
For the drug to be safe for human use, it should be free from impurities. Limit test is used to find out whether impurities are in permissible limit or not. In the pharmaceutical industry; quality control analysts must deal with raw material, side reactions, accelerate stability testing, storage conditions, and packaging material for the drugs etc. Performing a limit test gives an idea about the purity of chemicals (in the case of raw materials) or their stability (in the case of stability studies). In this experiment, students will perform a limit test for chloride present in common laboratory reagents that will help in testing the quality of drugs.

3. Practical Outcomes
After completion of this practical, the students will be able to:

PrO	Practical Outcomes	Mapped CO	BTL
1	Explain the principle and reaction involved in the limit test for chloride.	CO1-3	2
2	Prepare the reagents required for the limit test for chloride.	CO1-3	3
3	Perform the limit test for chloride.	CO1-3	4
4	Observe and compare the opalescence formed while performing the limit test.	CO1-3	4
5	Follow cleanliness, safety, and ethical practices.	CO1-3	5
6	Demonstrate working as a leader or team member.	CO1-3	5

4. Relevant Theoretical Background
Limit test for chlorides depends upon the interaction of chlorides with silver nitrate in presence of dilute nitric acid. This interaction of chloride ion with silver nitrate results in precipitation of chlorides as silver chloride. Silver chloride appears as opalescence only when it is present in very small quantities. As chlorides typically exist as impurities in pharmaceuticals in small quantities, the opalescence of silver chloride serves as a comparison. This comparison is made under standardized illumination conditions with the opalescence observed in a standard Nesssler's cylinder.



5. Requirements
Glasswares: Nessler's cylinder (50 mL), Measuring cylinder (5, 10 and 20 mL), Pipette (1 & 2 mL), Beaker (100 mL), Dropper, Test tube stand, Volumetric flask (100, 1000 mL).

Chemicals: Sodium chloride, Silver nitrate, Nitric acid, Aspirin, Sodium bicarbonate, Sodium acetate.

Reagents

a. **Chloride Standard Solution (25 ppm Cl):** Dilute 5 mL of a 0.0824% w/v solution of sodium chloride to 100 mL with distilled water

b. **0.1 M Silver Nitrate:** Dissolve 17.0 g of silver nitrate in sufficient water to 1000 mL using a volumetric flask.

c. **Dilute Nitric acid (5% v/v):** Measure 5.00 mL of the concentrated HNO_3, using a measurement cylinder or pipette and then, dilute to 100 mL with water using a volumetric flask.

6. Requirements used

_____

_____

7. Procedure

Prepare standard solution (A) and test solution (B) as mentioned in the following table and compare opalescence against dark (black) background.

Standard Solution (A)	Test Solution (B)		
	Aspirin	**Sodium bicarbonate**	**Sodium acetate**
a) In Nessler's cylinder, add 10 mL of chloride standard solution (25 ppm Cl). b) Add 5 mL of water. c) Add 10 mL of dil. Nitric acid. d)Dilute to 50 mL mark of Nessler's cylinder with distilled water. e) Add 1 mL of 0.1 M silver nitrate solution. f) Stir immediately with a glass rod and allow to stand for 5 min, protected from light.	a) Boil 1.75 g of aspirin with 75 mL of water for 5 min, add sufficient water to restore volume to 75 mL. Cool and filter. b) Transfer 25 mL of this filtrate to Nessler's cylinder. c) Add 10 mL of dil. Nitric acid. d)Dilute to 50 mL mark of Nessler's cylinder with distilled water. e) Add 1 mL of 0.1 M silver nitrate solution. f) Stir immediately with a glass rod and allow to stand for 5 min, protected from light. g) Observe transversely, the opalescence in test and standard against dark (black) background.	a) Dissolve 1.25 g of sodium bicarbonate in 15 mL distilled water and 2 ml of nitric acid. b) Transfer to Nessler's cylinder. c) Add 10 mL of dil. Nitric acid. d)Dilute to 50 mL mark of Nessler's cylinder with distilled water. e) Add 1 mL of 0.1 M silver nitrate solution. f) Stir immediately with a glass rod and allow to stand for 5 min, protected from light. g) Observe transversely, the opalescence in test and standard against dark (black) background.	a) Dissolve 10 g in sufficient carbon dioxide free water, adjust volume to 100 mL. b) Transfer 10 mL of the above solution in Nessler's cylinder. c) Add 10 mL of dil. Nitric acid. d)Dilute to 50 mL mark of Nessler's cylinder with distilled water. e) Add 1 mL of 0.1 M silver nitrate solution. f) Stir immediately with a glass rod and allow to stand for 5 min, protected from light. g) Observe transversely, the opalescence in test and standard against dark (black) background.

8. Precautions

a. Glass apparatus used for the limit test should be dried and cleaned.
b. Only distilled water should be used for performing limit tests
c. Do not suck acid or other chemicals by mouth, use pipette aid or suction bulb.
d. Protect silver chloride from light.

9. Observations

a. The opalescence produced by the test solution of aspirin was_____ (more/less/same) intense than that of standard solution.
b. The opalescence produced by the test solution of sodium bicarbonate was_____ (more/less/same) intense than that of standard solution.
c. The opalescence produced by the test solution of sodium acetate was _____ (more/less/same) intense than that of standard solution.

10. Result

a. The given sample of aspirin _____ (passes / doesn't pass) the limit test for chloride as per IP-2022.
b. The given sample of sodium bicarbonate _____ (passes / doesn't pass) the limit test for chloride as per IP-2022.
c. The given sample of sodium acetate _____ (passes / doesn't pass) the limit test for chloride as per IP-2022.

11. Conclusion

The limit test for chloride was performed on a given sample(s) of _____ as per IP 2022.

12. References

a. Indian Pharmacopoeia 2022.

13. Practical Related Questions

a. Define impurity and Limit test.
b. Describe the precautions while handling concentrated nitric acid.
c. Explain principle of limit test for chloride with reaction.
d. Why is nitric acid used in the chloride limit test? Give the reason.
e. State the difference between concentrated nitric acid and fuming nitric acid.

(*Space for Answers*)

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14. Assessment Scheme

Particular	Understanding the basic concept (Intellectual skill)	Activities (Motor skill)	Cleanliness & Handling (Affective domain)	Viva-voce / Answers Written	Total	Signature of teacher
Marks Obtained						
Max Marks	**02**	**05**	**01**	**02**	**10**	

Experiment No. 3

Limit Test for Sulphate

1. Aim

To perform and report the limit test for sulphate on the given samples as per IP-2022.

2. Practical Significance

For the drug to be safe for human use, it should be free from impurities. A limit test is used to find out whether impurities are in permissible limit or not. In the pharmaceutical industry; quality control analysts must deal with raw materials, side reactions, accelerate stability testing, storage conditions, and packaging material for the drugs etc. Performing a limit test gives an idea about the purity of chemicals (in the case of raw materials) or their stability (in the case of stability studies). In this experiment, students will perform a limit test for sulphate present in common laboratory reagents that will help in testing the quality of drugs.

3. Practical Outcomes

After completion of this practical, the students will be able to:

PrO	Practical Outcomes	Mapped CO	BTL
1	Explain the principle and reaction involved in the limit test for sulphate.	CO1-3	2
2	Prepare the reagents required for the limit test for sulphate.	CO1-3	3
3	Perform the limit test for sulphate.	CO1-3	4
4	Observe and compare the opalescence formed while performing the limit test.	CO1-3	4
5	Follow cleanliness, safety, and ethical practices.	CO1-3	5
6	Demonstrate working as a leader or team member.	CO1-3	5

4. Relevant Theoretical Background

The limit test serves the purpose of identifying sulphate impurities within compounds. When sulphate ions are present, they react with barium chloride in the presence of alcohol and potassium sulphate, resulting in turbidity. This turbidity arises from the formation of barium sulphate, which precipitates completely due to seeding. Alcohol, in small amounts, prevents the supersaturation of barium sulphate, ensuring the production of uniform turbidity. Potassium sulphate enhances the sensitivity of this test. Additionally, the presence of hydrochloric acid and acetic acid impacts acidity, facilitating the formation of insoluble barium sulphate and thus contributing to turbidity.

$$\underset{\text{Potassium sulphate}}{K_2SO_4} + \underset{\text{Barium chloride}}{BaCl_2} \xrightarrow{CH_3COOH} \underset{\text{Barium sulphate}}{BaSO_4\downarrow} + 2KCl$$

$$\underset{\text{Sulphate ion (Soluble)}}{SO_4^{2-}} + \underset{\text{Barium chloride}}{BaCl_2} \xrightarrow{CH_3COOH} \underset{\text{Barium sulphate}}{BaSO_4\downarrow} + 2Cl^-$$

5. Requirements

Glasswares: Nessler's cylinder (50 mL), Measuring cylinder (5 mL), Pipette (1 & 2 mL), Beaker (100 mL), Dropper, Test tube stand, Volumetric flask (25, 100 & 500 mL).

Chemicals: Ethanol, Potassium sulphate, Acetic acid, Barium chloride, Aspirin, Hydrochloric acid, Sodium bicarbonate, Citric acid

Reagents

a. **Ethanol (30 %):** 150 mL of ethanol is diluted with distilled water to produce 500 mL in a volumetric flask.

b. **Ethanolic Sulphate Standard Solution (10 ppm SO_4^{2-}):** Prepare 0.181% w/v solution of potassium sulphate in ethanol (30%). Dilute 1 mL of this solution of potassium sulphate to 100 mL with ethanol (30%).

c. **5 M acetic acid:** Measure 28.5 mL acetic acid, transfer in a volumetric flask, and add sufficient distilled water to produce 100 mL.

d. **25% w/v Barium chloride solution:** Weigh 6.25 g of barium chloride dissolve in sufficient distilled water to produce 25 mL.

e. **Standard Potassium sulphate Solution:** Accurately weigh 0.1089 g of K_2SO_4, was taken and the volume was made up to 100 mL with water.

6. Requirements used

_____

_____

7. Procedure

Prepare standard solution (A) and test solution (B) as mentioned in the following table and compare opalescence against dark (black) background.

Standard Solution (A)	**Test Solution (B)**		
	Aspirin	**Sodium bicarbonate**	**Citric acid**
a) In Nessler's cylinder, add 2 mL of potassium sulphate standard solution (20 ppm) and then add 1 mL 25% w/v solution of barium chloride. b) Then add 1.5 mL of ethanolic sulphate standard solution mix and allow to stand for 1 minute. c) Add 0.15 mL of 5 M acetic acid. d)Sufficient water shall be added to	a) In Nessler's cylinder add 1 mL 25% solution of barium chloride, then add 1.5 mL of ethanolic sulphate standard solution. Mix and allow to stand for 1 min. b) In a beaker, boil 1.75 g of aspirin with 75 mL of water for 5 min, add sufficient water to restore volume to 75 mL. c) Neutralize the solution with HCl and Transfer 10 mL of this filtrate to Nessler's cylinder.	a) In Nessler's cylinder add 1 mL 25% solution of barium chloride, then add 1.5 mL of ethanolic sulphate standard solution. Mix and allow to stand for 1 min. b) In a beaker, add 1.0 g sodium bicarbonate, add 10 mL of distilled water, neutralize this solution with HCl and then add 15 mL distilled water. Transfer this solution to Nessler's cylinder.	a) In Nessler's cylinder, add 1 mL 25% w/v solution of barium chloride, then add 1.5 mL of ethanolic sulphate standard solution mix and allow to stand for 1 min. b) In another beaker, add 1.0 g of citric acid, add 15 ml of water, neutralize with HCl. d) Transfer this solution in Nessler's cylinder and the add 0.15 mL of 5 M acetic acid.

produce 50 mL, stir with a glass rod and allow it to stand for 5 minutes.	d) Add 0.15 mL of 5M acetic acid. Add sufficient water to make up 50 mL volume. e) Stir with a glass rod and allow to stand for 5 min. f) Observe transversely, the opalescence in test and standard against dark (black) background.	d) Add 0.15 mL of 5 M acetic acid. e) Sufficient water shall be added to produce 50 mL, stir with a glass rod and allow it to stand for 5 minutes. f) Observe transversely, the opalescence in test and standard against dark (black) background.	e) Sufficient water shall be added to produce 50 mL, stir with a glass rod and allow it to stand for 5 minutes. f) Observe transversely, the opalescence in test and standard against dark (black) background.

8. Precautions

Glass apparatus used for the limit test should be dried and cleaned.
Only distilled water should be used for performing limit tests.
Different glass rods should be used for test and standard solutions.
Do not suck acid or other chemicals by mouth, use a pipette aid or suction bulb.
When mixing acid and water, always add concentrated acid to water dropwise with stirring.

9. Observations

a. The opalescence produced by the test solution of aspirin was_____ (more/less/same) intense than that of standard solution.
b. The opalescence produced by the test solution of sodium bicarbonate was_____ (more/less/same) intense than that of standard solution.
c. The opalescence produced by the test solution of citric acid was _____ (more/less/same) intense than that of standard solution.

10. Result

a. The given sample of aspirin _____ (passes / doesn't pass) the limit test for sulphate as per IP-2022.
b. The given sample of sodium bicarbonate _____ (passes / doesn't pass) the limit test for sulphate as per IP-2022.
c. The given sample of citric acid _____ (passes / doesn't pass) the limit test for sulphate as per IP-2022.

11. Conclusion

The limit test for sulphate was performed on a given sample(s) of _____ as per IP 2022.

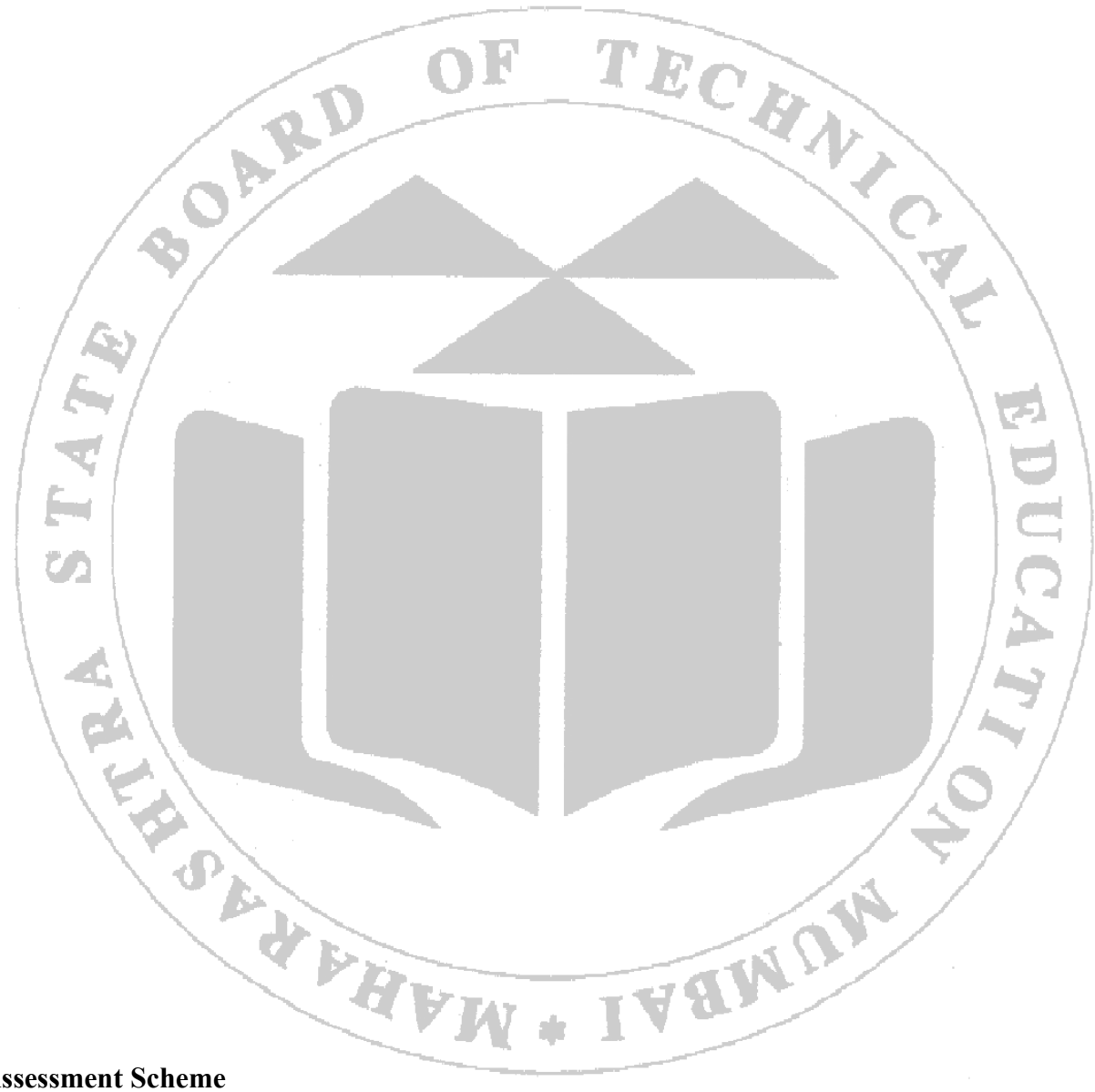
12. Reference

a. Indian Pharmacopoeia 2022.

13. Practical Related Questions

a. State the meaning of supersaturation.
b. What is the role of alcohol and hydrochloric acid in this limit test?
c. Explain the principle of limit test for Sulphate with reaction.
d. Write the procedure for the preparation of a standard solution for the limit test of sulphate.
e. Write the procedure for the preparation of 500 mL 5 M acetic acid.

(*Space for Answers*)



14. Assessment Scheme

Particular	Understanding the basic concept (Intellectual skill)	Activities (Motor skill)	Cleanliness & Handling (Affective domain)	Viva-voce / Answers Written	Total	Signature of teacher
Marks Obtained						
Max Marks	**02**	**05**	**01**	**02**	**10**	

Experiment No. 4

Limit Test for Iron

1. Aim

To perform and report the limit test for iron on the given samples as per IP-2022.

2. Practical Significance

For the drug to be safe for human use, it should be free from impurities. Limit test is used to find out whether impurities are in permissible limit or not. In the pharmaceutical industry; quality control analysts must deal with raw material, side reactions, accelerate stability testing, storage conditions and packaging material for the drugs etc. Performing a limit test gives an idea about the purity of chemicals (in case of raw materials) or its stability (in case of stability studies). In this experiment students will perform a limit test for iron present in common laboratory reagents that will help in testing the quality of drugs.

3. Practical Outcomes

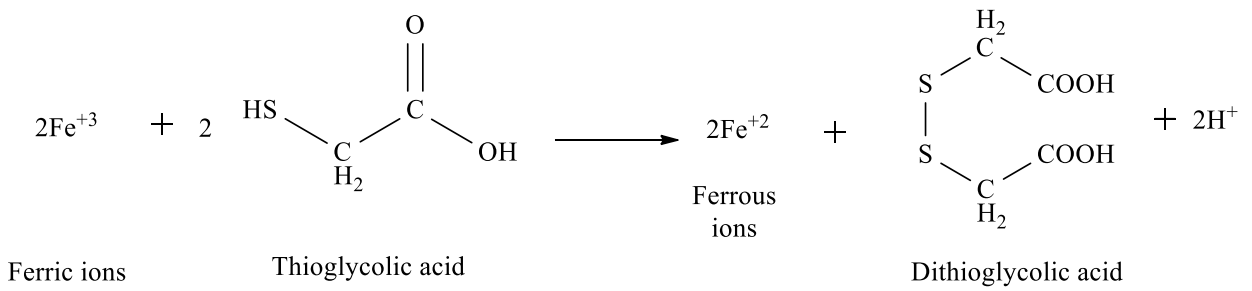
After completion of this practical, the students will be able to:

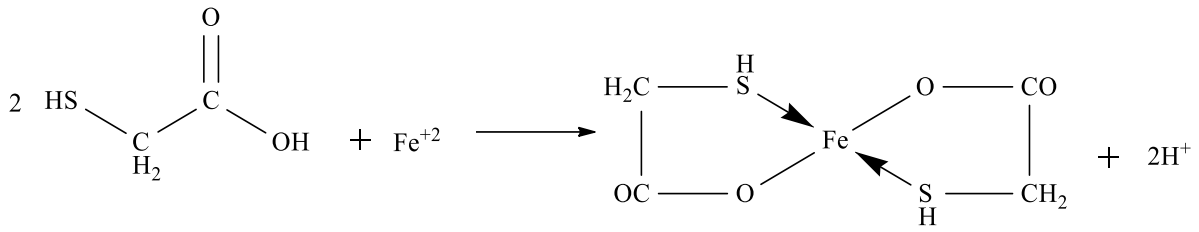
PrO	Practical Outcomes	Mapped CO	BTL
1	Explain the principle and reaction involved in the limit test for iron.	CO1-3	2
2	Prepare the reagents required for the limit test for iron.	CO1-3	3
3	Perform the limit test for iron.	CO1-3	4
4	Observe and compare the colour intensity formed while performing the limit test.	CO1-3	4
5	Follow cleanliness, safety, and ethical practices.	CO1-3	5
6	Demonstrate working as a leader or team member.	CO1-3	5

4. Relevant Theoretical Background

The limit test for iron is based on the interaction of iron with thioglycolic acid in the presence of citric acid and ammonia solution. Iron forms a purple-coloured ferrous thioglycolate complex. The original state of iron is insignificant, as thioglycolic acid reduces ferric ions (Fe^{3+}) to ferrous ions (Fe^{2+}). The ferrous ions then react with thioglycolic acid to form a coordination compound, i.e., the ferrous thioglycolate complex. This complex produces a purple colour only in an alkaline medium, so ammonia solution is used. Citric acid is used to prevent the precipitation of iron with ammonia; it forms an ammonium citrate buffer and keeps iron in solution by forming soluble complexes with iron.

Reaction





Thioglycolic acid — Ferrous ions — Ferous thioglycolate Complex

5. Requirements

Glasswares: Nessler's cylinder (50 mL), Measuring cylinder (5 mL), Pipette (1 & 2 mL), Beaker (100 mL), Dropper, Test tube stand, Volumetric flask (100, 500 & 1000mL).

Chemicals: Sulphuric acid, Ferric ammonium sulphate, Citric acid, sodium chloride, Sodium acetate, Calcium chloride, Thioglycolic acid, Ammonia solution, Hydrochloric acid.

Reagents

a. **0.05 M (0.1 N) sulphuric acid:** Add slowly, with stirring, 1.39 mL of sulphuric acid to about 500 mL of distilled water.

b. **Iron standard solution (20 ppm Fe):** In a volumetric flask add 0.1728 g ferric ammonium sulphate, dissolve 10 mL in 0.05 M sulphuric acid, and adjust the volume to 1000 mL with water This solution contains iron in a ferric state.

c. **20 % iron-free citric acid solution:** Dissolve 20 g of iron-free citric acid in 100 mL distilled water.

6. Requirements used

_____

_____

7. Procedure

Prepare standard solution (A) and test solution (B) as mentioned in the following table and compare opalescence against dark (black) background.

Standard Solution (A)	**Test Solution (B)**		
	Sodium chloride	**Sodium acetate**	**Calcium chloride**
a) In Nessler's cylinder, add 2 mL of iron standard solution (20 ppm Fe) and then add 2 mL 20% w/v solution of iron-free citric acid b) Then add 0.1 mL of thioglycolic acid, mix well, and	a) In Nessler's cylinder add 2 g of sodium chloride and dissolve in 20 mL of water. b) Add 2 mL 20% w/v solution of iron-free citric acid. c) Then add 0.1 mL of thioglycolic acid, mix well, and make alkaline with iron-free ammonia solution.	a) In Nessler's cylinder add 2 g of sodium acetate dissolved in 20 mL carbon dioxide-free water. b) Add 2 mL 20% w/v solution of iron-free citric acid c) Then add 0.1 mL of thioglycolic acid, mix well, and make alkaline with iron-free ammonia solution.	a) In Nessler's cylinder, add 2 g of calcium chloride dissolved in 0.5 mL HCl and 25 mL distilled water. b) Add 2 mL 20% w/v solution of iron-free citric acid c) Then add 0.1 mL of thioglycolic acid, mix well, and make alkaline with iron-free ammonia solution.

make alkaline with iron-free ammonia solution. c) Dilute to 50 mL with distilled water and allow to stand for 5 minutes.	d) Dilute to 50 mL with distilled water and allow to stand for 5 minutes. e) Observe transversely, the colour intensity in test and standard against a white background and compare with the standard.	d) Dilute to 50 mL with distilled water and allow to stand for 5 minutes. e) Observe transversely, the colour intensity in test and standard against a white background and compare with the standard.	d) Dilute to 50 mL with distilled water and allow to stand for 5 minutes. e) Observe transversely, the colour intensity in test and standard against a white background and compare with the standard.

8. Precautions

a. Glass apparatus used for the limit test should be dried and cleaned.
b. Only distilled water should be used for performing limit tests
c. Different glass rods should be used for test and standard solutions.
d. Do not suck acid or other chemicals by mouth, use pipette aid or suction bulb.
e. When mixing acid and water, always add concentrated acid to water dropwise with stirring.

9. Observations

a. The colour intensity produced by the test solution of sodium chloride was _____ (more/less/same) intense than that of the standard solution.
b. The colour intensity produced by the test solution of sodium acetate was _____ (more/less/same) intense than that of the standard solution.
c. The colour intensity produced by the test solution of calcium chloride was _____ (more/less/same) intense than that of the standard solution.

10. Result

a. The given sample of sodium chloride _____ (passes / doesn't pass) the limit test for iron as per IP-2022.
b. The given sample of sodium acetate _____ (passes/doesn't pass) the limit test for iron as per IP-2022.
c. The given sample of calcium chloride _____ (passes/doesn't pass) the limit test for iron as per IP-2022.

11. Conclusion

The limit test for iron was performed on a given sample(s) of _____ as per IP 2022.

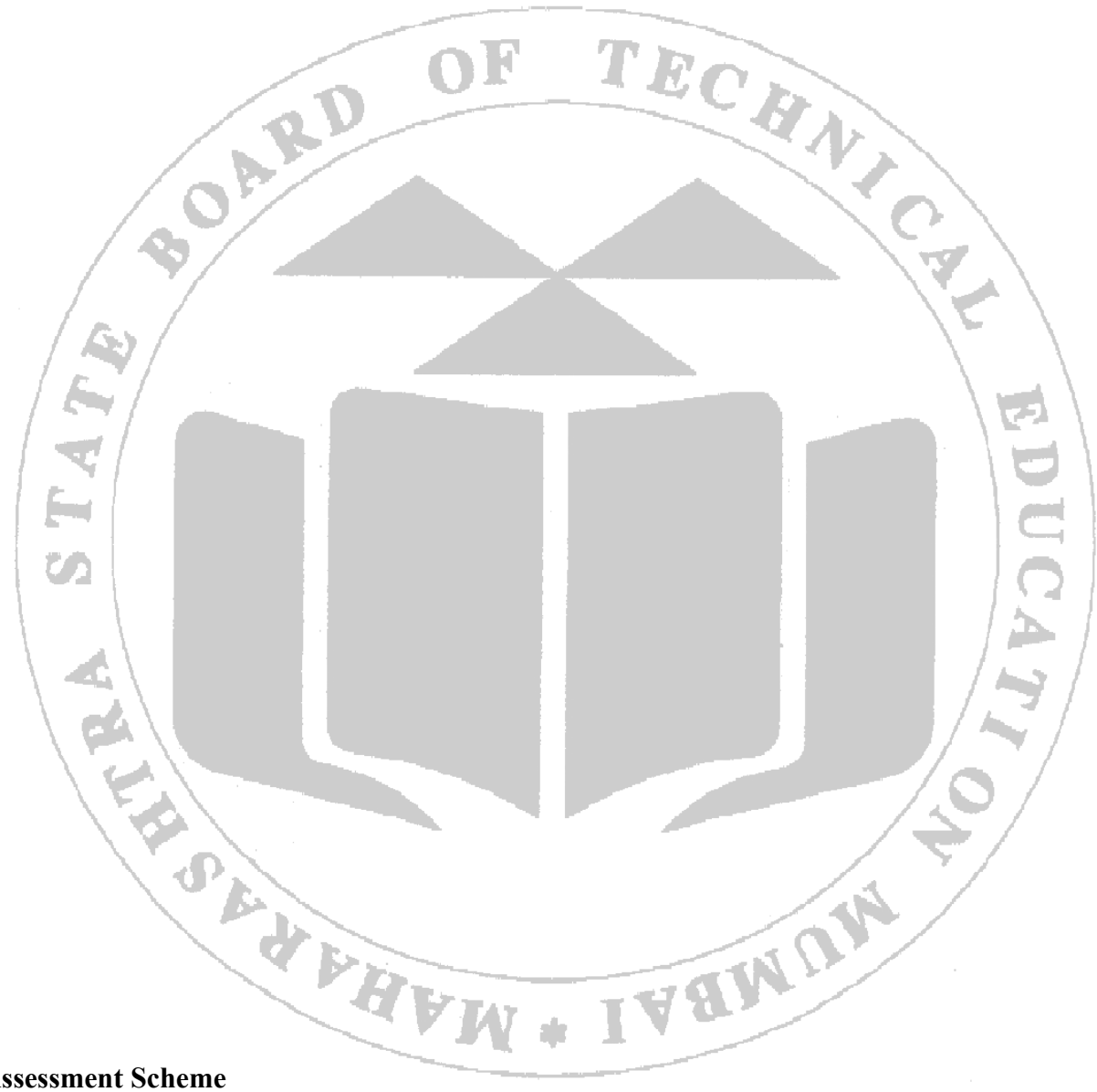
12. Reference

a. Indian Pharmacopoeia 2022.

13. Practical Related Questions

a. State the purpose of the limit test for iron.
b. Describe significance of citric acid and ammonia in the limit test for iron.
c. Explain the role of thioglycolic acid in the limit test for iron.
d. Write the procedure for preparation of standard solution of iron (20 ppm Fe).
e. What is the principle of limit test for iron?
f. Explain reaction and principle involved in the limit test for iron.

(*Space for Answers*)



14. Assessment Scheme

Particular	Understanding the basic concept (Intellectual skill)	Activities (Motor skill)	Cleanliness & Handling (Affective domain)	Viva-voce / Answers Written	Total	Signature of teacher
Marks Obtained						
Max Marks	**02**	**05**	**01**	**02**	**10**	

Experiment No. 5

Limit Test for Heavy Metals

1. Aim

To perform and report the limit test for heavy metals on the given samples as per IP-2022.

2. Practical Significance

For the drug to be safe for human use, it should be free from impurities. Limit test is used to find out whether impurities are in permissible limit or not. In the pharmaceutical industry; quality control analysts must deal with raw material, side reactions, accelerate stability testing, storage conditions and packaging material for the drugs etc. Performing a limit test gives an idea about the purity of chemicals (in case of raw materials) or its stability (in case of stability studies). In this experiment students will perform a limit test for heavy metals present in common laboratory reagents that will help in testing the quality of drugs.

3. Practical Outcomes

After completion of this practical, the students will be able to:

PrO	Practical Outcomes	Mapped CO	BTL
1	Explain the principle and reaction involved in the limit test for heavy metals.	CO1-3	2
2	Prepare the reagents required for the limit test for heavy metals.	CO1-3	3
3	Perform the limit test for heavy metals.	CO1-3	4
4	Observe and compare the opalescence formed while performing the limit test.	CO1-3	4
5	Follow cleanliness, safety, and ethical practices.	CO1-3	5
6	Demonstrate working as a leader or team member.	CO1-3	5

4. Relevant Theoretical Background

The limit test for heavy metals is based on the reaction between hydrogen sulphide and specific heavy metals like iron, lead, copper, nickel, and cobalt under acidic conditions, resulting in the formation of metal sulphides. These sulphides are dispersed in a colloidal state, imparting a brown coloration. The concentration of heavy metals in the substance is expressed as 'Parts of Lead per million parts of the substance' to signify the limit test outcome.

$$\text{Metal} + H_2S \xrightarrow[\text{pH 3-4}]{\text{dil } CH_3COOH} \text{Metal Sulphide} + H_2\uparrow$$

5. Requirements

Glassware: Nessler's cylinder (50 mL), Measuring cylinder (5, 10 mL), Pipette (1 & 2 mL), Beaker (100 mL), Dropper, Test tube stand, Volumetric flask (10, 25, 100 & 500 mL).

Chemicals: Lead nitrate, Nitric acid, Dil. Acetic acid, Dil. ammonia solution, Hydrogen sulphide solution, pH paper, Citric acid, Sodium chloride, Ascorbic acid.

Reagents

a. **Lead standard solution (20 ppm Pb):** First prepare a lead standard solution (0.1% Pb), for the preparation dissolve 0.4 g lead nitrate in 50 mL water already containing 2 mL of nitric acid. Then, add sufficient water to produce 250 mL in a volumetric flask. Pipette out 10 mL

of this solution and dilute to 100 mL with water in a volumetric flask (lead standard solution 100 ppm Pb). Take 10 mL of this 100 ppm Pb solution and dilute 50 ml in a volumetric flask with water to get a Lead standard solution of 20 ppm Pb.

6. Requirements used

_____

_____

7. Procedure

Prepare standard solution (A) and test solution (B) as mentioned in the following table and compare opalescence.

Standard Solution Method A as per IP 2022	**Test Solution (B)**		
	Citric acid	**Sodium chloride**	**Ascorbic acid**
a) In Nessler's cylinder, add 1.0 mL of lead standard solution (20 ppm Pb). b) Adjust volume to 25 mL with water. c) Adjust pH between 3.0 and 4.0, with dilute acetic acid or dilute ammonia solution. d) Then, dilute with water to about 35 mL and mix. e) Add 10 mL of freshly prepared H_2S solution and stir with a glass rod. f) Dilute to 50 mL with water, allow to stand for 5 minutes, and view downwards over a white surface.	a) In Nessler's cylinder, dissolve 2 g of citric acid in 10 mL of water, 5 mL of dilute hydrochloric acid and then add sufficient water to produce 25 mL. b) Adjust pH between 3.0 and 4.0, with dilute acetic acid or dilute ammonia solution. c) Then, dilute with water to about 35 mL and mix. d) Add 10 mL of freshly prepared H_2S solution and stir with a glass rod. e) Dilute to 50 mL with water, allow to stand for 5 minutes, and view downwards over a white surface, and compare with that of standard.	a) In Nessler's cylinder add 4 g of sodium chloride then add 2mL of dilute acetic acid. b) Mix well and add sufficient water, to produce 25 mL. c) Adjust pH between 3.0 and 4.0, with dilute acetic acid or dilute ammonia solution. d) Then, dilute with water to about 35 mL and mix. e) Add 10 mL of freshly prepared H_2S solution and stir with a glass rod. f) Dilute to 50 mL with water, allow to stand for 5 minutes, and view downwards over a white surface, and compare with that of standard.	a) In Nessler's cylinder, add 1 g of ascorbic acid, add 25 mL of water to dissolve. b) Adjust pH between 3.0 and 4.0, with dilute acetic acid or dilute ammonia solution. c) Then, dilute with water to about 35 mL and mix. d) Add 10 mL of freshly prepared H_2S solution and stir with a glass rod. e) Dilute to 50 mL with water, allow to stand for 5 minutes, and view downwards over a white surface, and compare with that of standard.

8. Precautions

a. Glass apparatus used for the limit test should be dried and cleaned.
b. Only distilled water should be used for performing limit tests
c. Different glass rods should be used for test and standard solutions.
d. Do not suck acid or other chemicals by mouth, use a pipette aid or suction bulb.
e. When mixing acid and water, always add concentrated acid to water dropwise with stirring.

9. Observations

a. The opalescence produced by the test solution of Citric acid was _____ (more/less/same) intense than that of the standard solution.
b. The opalescence produced by the test solution of Sodium chloride was _____ (more/less/same) intense than that of the standard solution.
c. The opalescence produced by the test solution of Ascorbic acid was _____ (more/less/same) intense than that of the standard solution.

10. Result

a. The given sample of Citric acid _____ (passes/doesn't pass) the limit test for heavy metals as per IP-2022.
b. The given sample of Sodium chloride _____ (passes/doesn't pass) the limit test for heavy metals as per IP-2022.
c. The given sample of Ascorbic acid _____ (passes/doesn't pass) the limit test for heavy metals as per IP-2022.

11. Conclusion

The limit test for heavy metal was performed on a given sample(s) of _____ as per IP 2022.

12. Reference

a. Indian Pharmacopoeia 2022.

13. Practical Related Questions

a. Why is there need for a limit test for heavy metals?
b. Explain the principle involved in the limit test for heavy metals.
c. Describe the procedure to prepare Lead standard solution 20 ppm Pb.
d. State the reason for using dilute acetic acid and dilute ammonia solution.
e. Is it possible to replace the H_2S solution by any other means? Justify with an example.
f. Write the chemical reaction involved in the limit test for heavy metals.

(*Space for Answers*)

14. Assessment Scheme

Particular	Understanding the basic concept (Intellectual skill)	Activities (Motor skill)	Cleanliness & Handling (Affective domain)	Viva-voce / Answers Written	Total	Signature of teacher
Marks Obtained						
Max Marks	**02**	**05**	**01**	**02**	**10**	

Experiment No. 6

Identification tests for Anions

1. Aim

To perform and report the identification test for the anions on the given sample.

2. Practical Significance

Ions play a vital role in the blood, body fluids, intracellular and extracellular environment. They are responsible for maintenance of acid-base balance and homeostasis. Ionization phenomenon, nature of solution and magnitude of ions plays a vital role in the various catalytic processes, reactions and their products in the industry. In the industry and pathology laboratory, chemists have to deal with the various solutions and their respective cations and anions. Various anions and cations can be identified in the blood, urine and different chemical compounds by means of chemical tests.

3. Practical Outcomes

After completion of this practical, the students will be able to:

PrO	Practical Outcomes	Mapped CO	BTL
1	Describe the reaction for the identification of anions.	CO3,5	2
2	Identify anions by performing a qualitative test.	CO3,5	4
3	Follow cleanliness, safety, and ethical practices.	CO3,5	5
4	Demonstrate working as a leader or team member.	CO3,5	5

4. Relevant Theoretical Background

Identification of anions in the solution is a type of qualitative analysis. Dissolution of acid, base and salt in the water gives anion and cation. Cation is positively charged ion while anion is negatively charged ion. The cation is formed by loss of electron(s) while the anion is formed by the gain of electron(s). Charge present on the anion and cation represent valency of the element. Various anions like carbonate (CO_3^{2-}), phosphate (PO_4^{3-}), sulphate (SO_4^{2-}), chloride (Cl^-), iodide (I^-) can be identified and confirmed by their respective chemical tests.

Reaction of Anions

1. Carbonate

$$CO_3^{2-} + 2HCl \longrightarrow 2Cl^- + 2H_2O + CO_2$$

$$CO_3^{2-} + BaCl_2 \xrightarrow{-2Cl^-} \underset{\text{Barium carbonate}}{BaCO_3} \xrightarrow{HCl} BaCl_2$$

2. Phosphate

$$2HPO_4^{2-} + \underset{\text{Silver ions}}{3Ag^+} \longrightarrow H_2PO_4^- + Ag_3PO_4$$

3. Sulphate

$$SO_4^{2-} + BaCl_2 \longrightarrow BaSO_4 + 2Cl^-$$

Barium sulphate

$$SO_4^{2-} + 2AgNO_3 \longrightarrow Ag_2SO_4 + NO_3^{2-}$$

Silver sulphate

4. Chromate or dichromate

$$CrO_4^{2-} + 2Ag^+ \longrightarrow Ag_2CrO_4$$

Silver ions — Silver chromate

$$Cr_2O_7^{2-} + 2Ag^+ \longrightarrow Ag_2Cr_2O_7$$

Silver ions — Silver dichromate

$$CrO_4^{2-} + BaCl_2 \longrightarrow BaCrO_4$$

Barium chromate

5. Acetate

$$CH_3COO^- + H_2SO_4 \longrightarrow CH_3COOH + HSO_4^-$$

$$CH_3COO^- + C_2H_5OH \xrightarrow[H_2SO_4]{} CH_3COOC_2H_5$$

Ethyl acetate

6. Halides

$$X^- + AgNO_3 \longrightarrow AgX + NO_3^{2-}$$

5. Requirements

Glasswares: Test tubes (10 mL×5), Beaker (100 mL), Dropper, Test tube stand, Test tube holder, Glass rod.

Chemicals: As per requirements.

6. Requirements used

_____

_____

7. Precautions

a. Use clean and dry test tubes.

b. Same glass rod/dropper/pipette should not be used because it will contaminate reagents.

8. Procedure

a. Clean the test tubes.
b. Perform chemical test and confirmatory test on samples as mentioned in the charts.
c. Sample solution can be prepared by dissolving sample (0.1-0.5 g) in water (5-10 mL).

Table for qualitative analysis

Identification and confirmatory test for carbonate (CO_3^{2-})

Identification test	Observation	Inference
1 mL sample solution + 2 mL conc HNO_3.	Effervescence of CO_2 gas	Carbonate (CO_3^{2-}) may be present
0.5 g of substance + heat the test tube, add 1 to 2 mL of dilute sulphuric acid or HCl and apply the cork with the delivery tube. Pass the evolved gas through the 2 mL lime water.	Formation of white turbidity or precipitate	Carbonate (CO_3^{2-}) may be present
To the 1 mL sample solution add 1 mL Barium chloride ($BaCl_2$) solution.	White precipitate appears	Carbonate (CO_3^{2-}) confirmed

Identification and confirmatory test for halides (Cl^- /Br^-/I^-)

Identification test	Observation	Inference
1 mL sample solution + 2 mL $AgNO_3$ solution.	White precipitate formed	Halides may be present
1 mL sample solution + 2 mL chloroform + chlorine water, shake well. (Chloroform layer will be at bottom)	$CHCl_3$ layer is colourless	Cl^- may be present
	$CHCl_3$ layer yellow/ brown	Br^- may be present
	$CHCl_3$ layer pink/violet	I^- May be present
C.T. for Cl^-		
1 mL sample solution + 2 mL lead acetate solution.	White precipitate	Cl^- confirmed
1 mL sample solution + 0.5 g MnO_2 + 1-2 mL H_2SO_4.	Green fumes produced, that changes moist blue litmus to red and then bleaches.	Cl^- confirmed
C.T. for Br^-		
1 mL sample solution + 2 mL lead acetate solution.	Brown precipitate	Br^- confirmed
1 mL sample solution + 0.5 g MnO_2 + 1-2 mL H_2SO_4 +Heat.	Brown Fumes	Br^- confirmed
C.T. for I^-		
1 mL sample solution + 2 mL lead acetate solution.	Yellow precipitate	I^-confirmed

Identification and confirmatory test for Sulphate (SO_4^{2-})

Identification test	Observation	Inference
1 mL sample solution + 2 mL barium nitrite solution.	White precipitate formed, insoluble in nitric acid	Sulphate (SO_4^{2-}) may be present
1 mL sample solution + 2 mL barium chloride solution.	White precipitate	Sulphate (SO_4^{2-}) Confirmed
1 mL concentrated sample solution + 1-2 mL $AgNO_3$ solution.	White crystalline precipitate	Sulphate (SO_4^{2-}) Confirmed

Identification and confirmatory test for Nitrates (NO_3^{2-})

Identification test	Observation	Inference
0.05 g solid salt + 1-2 mL conc sulphuric acid heat if necessary.	Yellow brown vapours of NO_2, pungent odour	Nitrates (NO_3^{2-}) may be present
Brown Ring test 0.05 g sample + 1.0 mL water to dissolve + add 1.0 mL sulphuric acid carefully alongside of test tube, mix and cool, carefully add 0.5 mL $FeSO_4$ solution without mixing.	Brown colour at the junction of two liquids	Nitrates (NO_3^{2-}) confirmed

Identification and confirmatory test for Acetate (CH_3COO^-)

Identification test	Observation	Inference
1 mL sample solution + 1 mL sulphuric acid + 2 mL ethanol heat.	Fruity smell of ethyl acetate	CH_3COO^- may be present
0.5 g of solid salt + 2-3 mL dil sulphuric acid solution, heat if necessary.	Vapours of acetic acid, with irritating smell	CH_3COO^- confirmed

Identification and confirmatory test for Phosphate (PO_4^{3-})

Identification test	Observation	Inference
1 mL sample solution + 1 mL silver nitrate solution.	Yellow precipitate of silver phosphate, readily soluble in nitric acid and ammonia	Phosphate (PO_4^{3-}) may be present
1 mL sample solution + 2-3 mL dilute nitric acid solution (acidify the solution) + add 2-3 mL ammonium molybdate solution slowly.	Yellow crystalline precipitate of ammonium phosphomolybdate appears	Phosphate (PO_4^{3-}) confirmed

Identification and confirmatory test for chromates (CrO_4^{2-}) or Dichromates ($Cr_2O_7^{2-}$)

Identification test	Observation	Inference
1 mL sample solution + 1 mL silver nitrate solution.	Brownish red precipitate of silver chromate produced, readily soluble in nitric acid and ammonia, insoluble in acetic acid	CrO_4^{2-} or $Cr_2O_7^{2-}$ may be present
1 mL sample solution + 2 mL barium chloride solution.	Yellow precipitate formed	CrO_4^{2-} or $Cr_2O_7^{2-}$ may be present
1 mL sample solution + 2 mL of conc sulphuric acid + pass H_2S gas.	Green colour produced	CrO_4^{2-} or $Cr_2O_7^{2-}$ confirmed
1 mL sample solution + 2 mL of conc sulphuric acid + 0.5 mL diphenyl carbazide solution.	Deep red colour formed	CrO_4^{2-} or $Cr_2O_7^{2-}$ confirmed

9. Observations

Sample No. _____.

Identification and confirmatory tests for anions

Observation table

Identification Test	Observation	Inference

Identification Test	Observation	Inference

10. Result

The given sample _____was found to contain _____anions.

11. Conclusion

The identification test of anion was performed on a given sample.

12. References

a. Indian Pharmacopoeia 2022.
b. A Laboratory Manual for Basic Chemistry (22102), Maharashtra State Board of Technical Education, Mumbai.

13. Practical Related Questions

a. Why is there a need for identification of anions?
b. Write reaction for chromate.
c. How are sulphate salts identified?
d. Write a confirmatory test for nitrates.
e. Write reaction for sulphate.

(*Space for Answers*)

14. Assessment Scheme

Particular	Understanding the basic concept (Intellectual skill)	Activities (Motor skill)	Cleanliness & Handling (Affective domain)	Viva-voce / Answers Written	Total	Signature of teacher
Marks Obtained						
Max Marks	**02**	**05**	**01**	**02**	**10**	

Experiment No. 7

Identification test for Cations

1. Aim

To perform and report the identification test for the cations on the given sample.

2. Practical Significance

Ions play a vital role in the blood, body fluids, intracellular and extracellular environment. They are responsible for maintenance of acid-base balance and homeostasis. Ionization phenomenon, nature of solution and magnitude of ions plays a vital role in the various catalytic processes, reactions and their products in the industry. In the industry and pathology laboratory, chemists have to deal with the various solutions and their respective cations and anions. Various anions and cations can be identified in the blood, urine and different chemical compounds by means of chemical tests.

3. Practical Outcomes

After completion of this practical, the students will be able to:

PrO	Practical Outcomes	Mapped CO	BTL
1	Describe the reaction for the identification of cations.	CO3,5	2
2	Identify cations by performing a qualitative test.	CO3,5	4
3	Follow cleanliness, safety, and ethical practices.	CO3,5	5
4	Demonstrate working as a leader or team member.	CO3,5	5

4. Relevant Theoretical Background

Identification of cations in the solution is a type of qualitative analysis. Dissolution of acid, base and salt in the water give anion and cation. Cation is positively charged ion while anion is negatively charged ion. The cation is formed by loss of electron(s) while the anion is formed by the gain of electron(s). The charge present on the anion and cation represent valency of the element.

Various cations like silver (Ag^{+}), Calcium (Ca^{2+}), Barium (Ba^{2+}), Ferric (Fe^{3+}), Ferrous (Fe^{2+}), Sodium (Na^{+}), potassium (K^{+}) can be identified and confirmed by their respective chemical tests.

5. Requirements

Glasswares: Test tubes (10 mL×5), Beaker (100 mL), Dropper, Test tube stand, Test tube holder, Glass rod**.**

Chemicals: As per requirements.

6. Requirements used

_____

_____

7. Precautions

a. Use clean and dry test tubes.
b. Same glass rod/dropper/pipette should not be used because it will contaminate reagents.

8. Procedure

a. Clean the test tubes.

b. Perform chemical test and confirmatory test on samples as mentioned in the charts.
c. Sample solution can be prepared by dissolving sample (0.1-0.5 g) in water (5-10 mL).
d. Add equal quantity of relevant reagents as per chart given below

Table for qualitative analysis

Identification of Cations

S. No.	Test	Observation	Inference
1.	O.S. + dil. HCl	White ppt.	I group present i.e. Pb^{2+} may be present.
		No ppt.	I group is absent
2.	O.S.+ dil. HCl + H_2S gas	ppt. obtained	II Group present
		1.Black ppt. of CuS	Cu^{2+} may be present
		2. Brown ppt. OF SnS	Sn^{2+} may be present
		3.Yellow ppt. of SnS_2	Sn^{4+} may be present
		No ppt.	II group is absent
3.	O.S.+ NH_4Cl (excess) +NH_4OH (till alkaline)	ppt. obtained	III A Group present.
		1.White gelatinous ppt of Al $(OH)_3$	Al^{3+} may be present.
		2. Dirty green ppt. of Fe $(OH)_2$	Fe^{2+} may be present
		3. Reddish brown ppt. of Fe $(OH)_3$	Fe^{3+} may be present
		4. Bluish green ppt. of Cr $(OH)_3$	Cr^{3+} may be present
		No ppt	III A group is absent
4.	O.S.+ NH_4Cl (excess) + NH_4OH (till alkaline) + H_2S gas	ppt obtained	III B Group present
		1. White ppt of ZnS	Zn^{2+} may be present
		2. Faint pink ppt of MnS	Mn^{2+} may be present
		3. Black ppt of NiS or CoS	Ni^{2+} or Co^{2+} may be present
		No ppt.	III B group is absent
	Above Black ppt. obtained + Conc.HNO_3	Green solution	Ni^{2+} present
		Blue Solution	Co^{2+} present
5.	O.S.+ NH_4Cl (excess) + NH_4OH (till alkaline) + $(NH_4)_2CO_3$	White ppt. of $CaCO_3$ or $BaCO_3$	IV group is present i.e. Ba^{2+} or Ca^{2+} may be Present
6.	O.S.+ K_2CrO_4	Yellow ppt.	Ba^{2+} may be present
		No ppt.	Ca^{2+} may be present

If all the above groups are absent then proceed for detection of Na^+, K^+ and NH_4^+

S. No.	Test	Observation	Inference
1.	O.S.+ NaOH (Boil)	Smell of ammonia gas or turns moist red litmus blue	NH_4^+ May be present
		No smell of ammonia, does not turn moist red litmus blue	Na^+ or K^+ may be present

S. No.	Test	Observation	Inference
2.	O.S.+ Sodium cobaltinitrite [fresh solution]	Yellow ppt.	K^+ may be present
		No ppt.	Na^+ may be present

Confirmatory Test (C.T.) for cations

C.T. for GROUP I cation

C. T. for Pb^{2+}

Sr. No.	Test	Observation	Inference
1.	O. S.+ dil. H_2SO_4	White ppt.	Pb^{2+} confirmed
2.	O.S.+ KI	Deep yellow ppt.	Pb^{2+} confirmed
3.	O.S.+ K_2CrO_4	Yellow ppt.	Pb^{2+} confirmed

C.T. For GROUP II cations

C.T. for Cu2+

Sr.No.	Test	Observation	Inference
1.	O.S. +K_4[Fe $(CN)_6$]	Chocolate red ppt.	Cu^{2+} Confirmed
2.	O.S.+ KI	Brown ppt.	Cu^{2+} Confirmed
3.	O.S.+ NaOH	Blue ppt.	Cu^{2+} Confirmed

C.T. for Sn^{2+}

Sr. No.	Test	Observation	Inference
1.	O.S.+ $HgCl_2$	White ppt. turns gray	Sn^{2+} Confirmed
2.	O. S. + NaOH	White ppt. soluble in excess of NaOH	Sn^{2+} Confirmed
3.	O.S.+ Iodine solution	Decolorization of iodine solution	Sn^{2+} Confirmed

C.T. for GROUP III A cations

C.T. For Al3+

Sr. No.	Test	Observation	Inference
1.	O. S.+ NaOH	White gelatinous ppt.	Al^{3+} Confirmed
2.	O.S.+ Ammonium acetate solution	No ppt in cold but gives white gelatinous ppt on boiling	Al^{3+} Confirmed
3.	O.S.+ NaH_2PO_4 (Monosodium phosphate)	White gelatinous ppt soluble in dil. HCl	Al^{3+} Confirmed

C.T. For Fe^{2+}

Sr. No.	Test	Observation	Inference
1.	O.S.+ $K_3[Fe(CN)_6]$	Deep Blue ppt.	Fe^{2+} Confirmed
2.	O.S.+ NaOH	Dirty green ppt.	Fe^{2+} Confirmed
3.	O.S.+ dil. H_2SO_4 + 1% $KMnO_4$ solution.	Pink colour of $KMnO_4$ decolorizes	Fe^{2+} Confirmed

C.T. For Fe^{3+}

Sr. No.	Test	Observation	Inference
1.	O.S.+$K_3[Fe(CN)_6]$	Deep Blue ppt.	Fe^{3+} Confirmed
2.	O.S. + NaOH	Reddish brown ppt.	Fe^{3+} Confirmed
3.	O.S. + Ammonium thiocyanate solution	Blood red ppt.	Fe^{3+} Confirmed

C.T. for Cr^{3+}

Sr. No.	Test	Observation	Inference
1.	O. S.+ NaOH	Bluish Green ppt.	Cr^{3+} Confirmed
2.	O.S. + PbO_2 + NaOH Boil, collect supernatant solution in another test tube and add acetic acid	Yellow ppt.	Cr^{3+} Confirmed

C.T. for Group III (B) cations

C.T. for Zn^{2+}

Sr. No.	Test	Observation	Inference
1.	O. S.+ NaOH	White ppt. insoluble in dil. HCl	Zn^{2+} Confirmed
2.	O.S.+NaH_2PO_4	White ppt.	Zn^{2+} Confirmed
3.	O.S.+$K_3[Fe(CN)_6]$	White ppt.	Zn^{2+} Confirmed

C.T. for Mn^{2+}

Sr. No.	Test	Observation	Inference
1.	O.S .+ NaOH	White ppt. soluble in excess of NaOH	Mn^{2+} Confirmed
2.	O.S. + NaOH + Br_2 water	Black ppt.	Mn^{2+} Confirmed
3.	O.S.+$K_3[Fe(CN)_6]$	Pinkish white ppt. soluble in dil. HCl	Mn^{2+} Confirmed

C.T. for Ni^{2+}

Sr. No.	Test	Observation	Inference
1.	O.S.+ NaOH + Br_2 Water	Black ppt.	Ni^{2+} Confirmed
2.	O.S.+NH_4OH	Pale green ppt, soluble in excess giving blue solution	Ni^{2+} Confirmed
3.	O.S. + Dimethylglyoxime	Scarlet red ppt.	Ni^{2+} Confirmed

C.T. for Co^{2+}

Sr. No.	Test	Observation	Inference
1.	**Test**	**Observation**	**Inference**
2.	O.S.+ NH_4OH	Blue ppt. turns brown in excess	Co^{2+} Confirmed
3.	O.S.+ Ammonium thiocvnate	Black ppt.	Co^{2+} Confirmed

Sr. No.	Test	Observation	Inference
	O.S.+ $K_3[Fe(CN)_6]$	Reddish ppt.	Co^{2+} Confirmed

C.T. for Group IV cations

C.T. for Ba^{2+}

Sr. No.	Test	Observation	Inference
1.	O.S.+ K_2CrO_4 (Potassium chromate)	Yellow ppt.	Ba^{2+} Confirmed
2.	O.S.+ Ammonium oxalate	White ppt.	Ba^{2+} Confirmed
3.	O.S. + dil.H_2SO_4	White ppt.	Ba^{2+} Confirmed

C.T. for Ca^{2+}

Sr. No.	Test	Observation	Inference
1.	O.S.+ K_2CrO_4(potassium chromate)	No ppt.	Ca^{2+} Confirmed
2.	O.S.+ Ammonium oxalate	White ppt. insoluble in acetic acid	Ca^{2+} Confirmed
3.	O. S. + NH_4Cl (crystals) + $K_3[Fe(CN)_6]$	White ppt.	Ca^{2+} Confirmed
4.	Flame Test	Brick Red coloured flame	Ca^{2+} Confirmed

C.T. for Group V cations

C.T. for Mg^{2+}

Sr. No.	Test	Observation	Inference
1.	O.S.+ NaOH	White ppt.	Mg^{2+} confirmed
2.	O.S.+ Hypoiodide solution	Reddish brown ppt.	Mg^{2+} confirmed

C.T. for NH_4^+

Sr. No.	Test	Observation	Inference
1.	O.S.+ Nessler's reagent	Brown ppt.	NH_4^+Confirmed
2.	O.S.+ Picric acid (alcoholic)	Yellow crystalline ppt.	NH_4^+ Confirmed

C.T. for K^+

Sr. No.	Test	Observation	Inference
1.	O.S. + Sodium cobaltinitrite Solution (freshly prepared)	Yellow ppt.	K^+ Confirmed
2.	O.S.+ Picric acid (alcoholic)	Yellow ppt.	K^+ Confirmed
3.	O.S.+ Perchloric acid	White ppt.	K^+ Confirmed

C.T. For Na^+

Sr. No.	Test	Observation	Inference
1.	O.S. + Sodium cobaltinitrite solution (freshly prepared)	Yellow ppt.	Na^+ Confirmed
2.	Flame test	Golden yellow flame	Na^+ Confirmed

*O.S. - Original water solution of given inorganic salt, ppt.- precipitate. dil- Dilute, Conc. - Concentrated, C.T. - Confirmatory test.

9. Observations

Sample No. _____.

Identification and confirmatory tests for cations.

Observation table

Identification Test	Observation	Inference

Identification Test	Observation	Inference

10. Result

The given sample _____was found to contain _____cations.

11. Conclusion

The identification test of cation was performed on a given sample.

12. References

a. Indian Pharmacopoeia 2022.

b. A laboratory manual for basic chemistry (22102), Maharashtra State Board of Technical Education, Mumbai.

13. Practical Related Questions

a. Why is there a need for identification of cations?
b. Describe reaction for identification and confirmation of Ca^{2+}.
c. Describe C. T. For Group IV cations.
d. How will you confirm K^{+}?

(*Space for Answers*)

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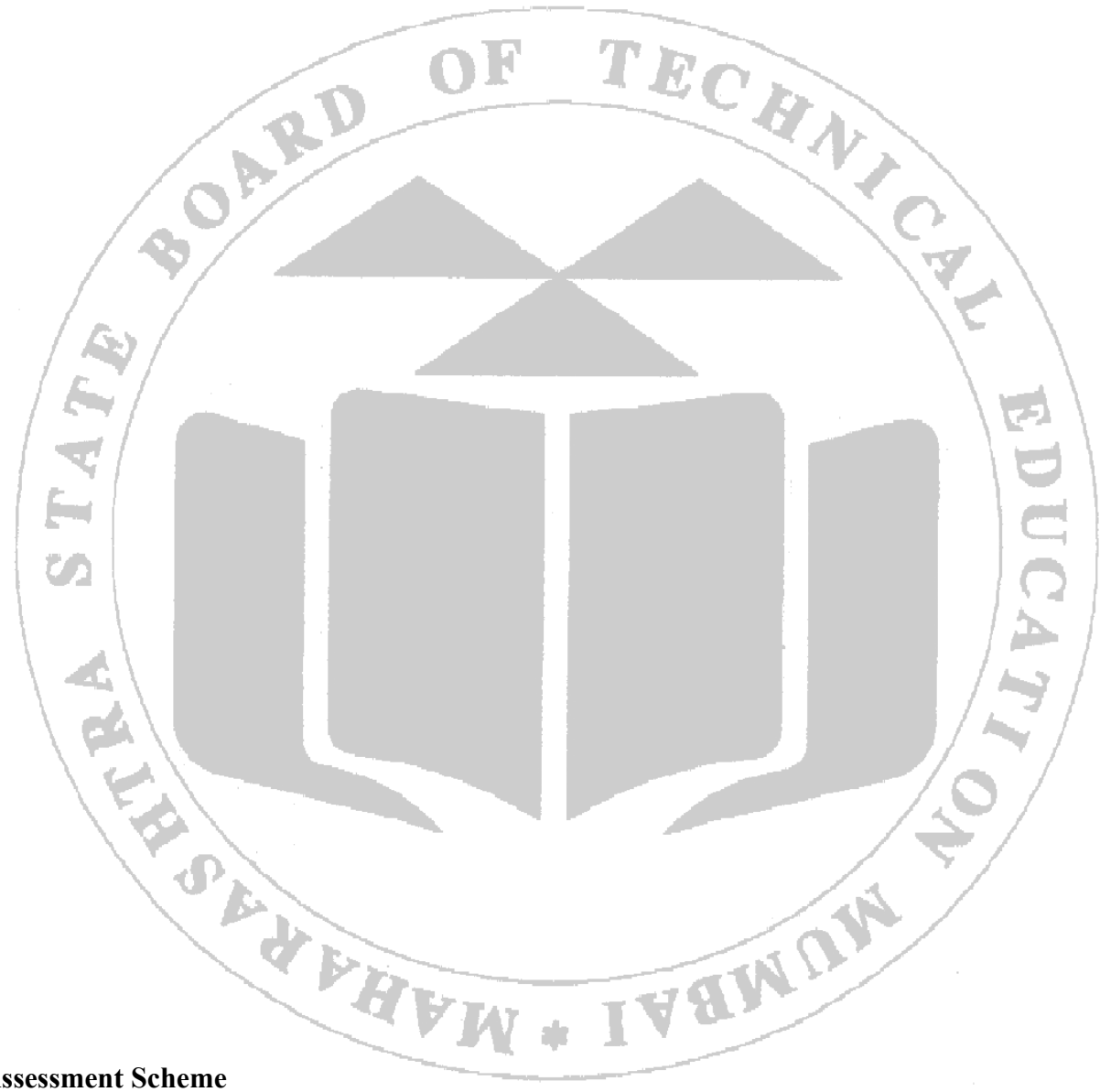


14. Assessment Scheme

Particular	Understanding the basic concept (Intellectual skill)	Activities (Motor skill)	Cleanliness & Handling (Affective domain)	Viva-voce / Answers Written	Total	Signature of teacher
Marks Obtained						
Max Marks	**02**	**05**	**01**	**02**	**10**	

Experiment No. 8
Preparation and standardization of Sodium hydroxide solution.

1. Aim
To prepare and standardize 1 M sodium hydroxide solution as per IP-2022.

2. Practical Significance
To carry out volumetric analysis, it is necessary for the chemist to be familiar with the methods for preparing solutions of various normality or molarity concentrations. The preparation and standardization of secondary standard solutions are critical steps in various analytical procedures. These processes ensure accuracy and reliability in quantitative chemical analysis. In this experiment, students will prepare and standardize a sodium hydroxide solution.

3. Practical Outcomes
After completion of this practical, the students will be able to:

PrO	Practical Outcomes	Mapped CO	BTL
1	Explain the principles and reactions involved in acid-base titration.	CO2,3	2
2	Prepare 1M sodium hydroxide solution.	CO2,3	3
3	Perform the standardization and calculate the exact molarity of the prepared solution.	CO2,3	4
4	Follow precision in weighing and precautions while handling the chemicals.	CO2,3	5
5	Demonstrate working as a leader or team member.	CO2,3	5

4. Relevant Theoretical Background

Acid-Base Titration

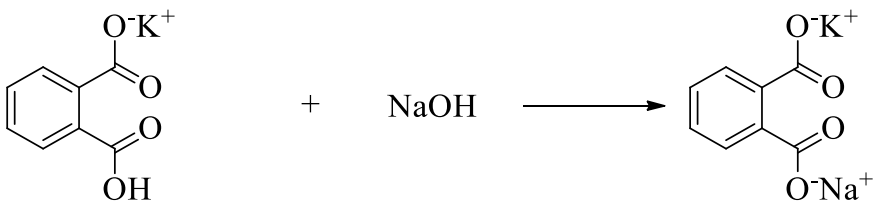
Acid-base titration is a precise and widely used method to determine the concentration of an unknown acid or base by reacting it with a standard solution of known concentration. The fundamental reaction in an acid-base titration is the neutralization reaction between the acid and the base. This can be represented by the general equation:

Acid + Base → Salt + Water

Principle

The principle of acid-base titration is based on the neutralization reaction between the hydrogen ions (H^+) from the acid and the hydroxide ions (OH^-) from the base.

In this experiment, Sodium hydroxide (base) is the secondary standard; hence, it is needed to standardize the prepared sodium hydroxide solution by titrating against primary standard acid. According to IP 2018, Sodium hydroxide is standardized by titration against primary standard potassium hydrogen phthalate(acid).



Potassium hydrogen phthalate (KHP)

5. Requirements

Glasswares: Burette (50 mL or 100 mL), Measuring cylinder (100 mL), Conical flask (250 mL), Dropper, Burette stand, Volumetric flask (500 mL).

Chemicals: Phenolphthalein indicator, Potassium hydrogen phthalate (KHP), Sodium hydroxide, Carbon dioxide free water.

6. Requirements used

_____

_____

7. Precautions

a. Adding water to solid sodium hydroxide generates heat, cool the flask in ice water.
b. Before filling the burette for the titration, rinse it with distilled water and then pre-rinse it with a portion of the titrant solution.
c. Remove air bubbles from the burette and adjust the reading to zero.

8. Procedure

Step I) Preparation of 1 M NaOH solution

Dissolve 42 g of NaOH in sufficient carbon-dioxide free water (500 mL), cool the flask, make up the volume to 1000 mL with distilled water in a volumetric flask. *(Sodium hydroxide is hygroscopic in nature. So, for the preparation of standard solution more than 1-gram equivalent i.e. little over 40 g of NaOH is weighted out.)*

Step II) Standardization of 1 M NaOH solution

a. Weigh accurately 5 g of pure and dried potassium hydrogen phthalate by the method of difference.
b. Transfer in dry conical flask and dissolve it in 75 mL of carbon-dioxide free water.
c. Add one drop of Phenolphthalein indicator.
d. Fill a clean burette with 1 M NaOH solution up to zero mark.
e. Place the flask below the burette, add slowly 1 M NaOH solution dropwise until the solution in the flask is faintly pink. Take burette reading.
f. Repeat this process for 2 more times.

(NaOH can also be standardized by titrating against succinic acid, oxalic acid, benzoic acid.)

9. Observations

Standardization of NaOH solution

a) Solution in burette: Prepared NaOH solution.

b) Contents of the conical flask: 5 g KHP + 75 mL of water + one drop of Phenolphthalein as an indicator.

c) End Point: Colourless to faint pink colour.

Observation table

Sr. No	Initial burette reading (mL)	Final burette reading (mL)	Difference (mL)	Mean burette reading (MBR) (mL)
1.				
2.				
3.				

Calculations

Factor calculation

1 molecule of NaOH reacts with 1 molecule of KHP

NaOH (40 g) ≅ KHP (204.23 g)

1000 mL of 1 M NaOH (40 g of NaOH dissolved in 1000 mL) = 204.23 g of KHP

Therefore,

1 mL of 1 M NaOH = 0.20423 g of KHP (Factor for standardization)

$$\text{Molarity} = \frac{\text{Weight of Potassium hydrogen phthalate}}{\text{MBR x Factor}}$$

$$m = \frac{5}{\text{MBR x } 0.20423} = \frac{24.48}{}$$

Therefore, calculated molarity of Sodium hydroxide = _____M

10. Result

The molarity of the prepared NaOH solution was found to be_____ M.

11. Conclusion

1 M NaOH solution was prepared and standardized as per the procedure given in IP 2022.

12. Reference

a. Indian Pharmacopoeia 2022.

13. Practical Related Questions

a. Calculate factor for standardization of 1 M NaOH.
b. Describe the principle involved in neutralization reaction.
c. Enlist precautions while handling solid NaOH.
d. Enlist any two primary standard and secondary standard compounds.
e. Write the reaction for NaOH standardization with KHP.

(*Space for Answers*)

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14. Assessment Scheme

Particular	Understanding the basic concept (Intellectual skill)	Activities (Motor skill)	Cleanliness & Handling (Affective domain)	Viva-voce / Answers Written	Total	Signature of teacher
Marks Obtained						
Max Marks	**02**	**05**	**01**	**02**	**10**	

Experiment No. 9
Preparation and standardization of Potassium permanganate solution

1. Aim
To prepare and standardize 0.02 M potassium permanganate solution as per IP-2022.

2. Practical Significance
To carry out volumetric analysis, it is necessary for the chemist to be familiar with the methods for preparing solutions of various normality or molarity concentrations. The preparation and standardization of secondary standard solutions are critical steps in various analytical procedures. These processes ensure accuracy and reliability in quantitative chemical analysis. In this experiment, students will prepare and standardize a potassium permanganate solution.

3. Practical Outcomes
After completion of this practical, the students will be able to:

PrO	Practical Outcomes	Mapped CO	BTL
1	Explain the principle and reaction involved in permanganometry.	CO2,3	2
2	Prepare 0.02 M potassium permanganate solution.	CO2,3	3
3	Perform the standardization and calculate the exact molarity of the prepared solution.	CO2,3	4
4	Follow precision in weighing and precautions while handling the chemicals.	CO2,3	5
5	Demonstrate working as a leader or team member.	CO2,3	5

4. Relevant Theoretical Background
Permanganometry is a type of redox titration in which potassium permanganate ($KMnO_4$) is used as the titrant. Potassium permanganate is a strong oxidizing agent and can be used to determine the concentration of reducing agents in a solution. Potassium permanganate is a powerful oxidizing agent, especially in acidic solutions, where it is reduced from Mn^{7+} to Mn^{2+}. The half-reaction for the reduction of potassium permanganate in acidic medium is:

$$\underset{\text{Permanganate ion}}{MnO_4^-} + 8H^+ + 5e^- \longrightarrow \underset{\text{Manganese ion}}{Mn^{2+}} + 4H_2O$$

$KMnO_4$ acts as its own indicator. In acidic solution, it is purple, and upon reduction, it becomes colorless. The disappearance of the purple color indicates the endpoint of the titration.

Official books such as IP 2022, mentions standardization of $KMnO_4$ solution by iodometric method, Potassium permanganate oxidizes KI into iodine in acidic media, the liberated iodine is titrated with sodium thiosulphate till yellow colour appears. At this stage starch mucilage is added as an indicator, the resulting blue colour is discharged on continuing the titration at the end point.

$$2KMnO_4 + 10KI + 8H_2SO_4 \longrightarrow 6K_2SO_4 + 2MnSO_4 + 5I_2\uparrow + 8H_2O$$

$$I_2 + \underset{\text{Sodium thiosulphate}}{2Na_2S_2O_3} \longrightarrow \underset{\text{Sodium tetrathionate}}{Na_2S_4O_6} + 2NaI$$

5. Requirements

Glasswares: Burette (50 mL or 100 mL), Measuring cylinder (100 mL), Conical flask (250 mL), Dropper, Burette stand, Volumetric flask (500 mL)

Chemicals: Potassium permanganate, Sodium thiosulphate, Potassium iodide, Sulphuric acid, Starch solution.

Reagents

a. **0.1 M sodium thiosulphate:** Dissolve 24.8 g of sodium thiosulphate and 0.2 g of sodium carbonate in sufficient carbon-dioxide free water to produce 1000 mL.

b. **1 M sulphuric acid:** Carefully add dropwise 54 mL of sulphuric acid to an equal volume of water and then dilute to 1000 mL with water.

6. Requirements used

_____

_____

7. Precautions

a. Use clean and dry glass apparatus.

b. Great care should be taken in handling potassium permanganate as dangerous explosions are liable to occur if it is brought into contact with readily oxidizable substances, either in solution or in the dry condition.

c. Ordinary distilled water likely to contain traces of reducing substances, that will act with potassium permanganate to form manganese dioxide, this will trigger auto decomposition of permanganate solution on standing. This is the reason for the heating solution, keeping it for two days and then filtration. The precipitated MnO_2 is removed by filtration.

8. Procedure

Step I) Preparation of 0.02 M $KMnO_4$ solution

a. Dissolve 3.16 g of potassium permanganate in 500 mL beaker containing water.

b. Stir thoroughly, breaking up the crystals with a glass rod.

c. Heat on a water bath for 1 hour. Allow to stand for two days and filter through glass wool.

d. Transfer solution in 1000 mL volumetric flask and make up the volume up to graduation mark with water, mix thoroughly with shaking.

Step II) Standardization of 0.02 M $KMnO_4$ solution

a. Rinse the burette with 0.1 M sodium thiosulphate solution and then, fill it up to zero mark with 0.1 M sodium thiosulphate.

b. Take 20.0 mL of prepared 0.02 M potassium permanganate solution in an iodine flask, add 2 g potassium iodide, acidify with 10 mL 1 M sulfuric acid solution, shake well.

c. Quantitative liberation of iodine occurs immediately. Titrate the iodine liberated in iodine flask by dropwise addition of 0.1 M sodium thiosulphate solution.

d. When the solution assumes yellow-green colour, add 3 mL of starch solution. The solution becomes blue in color.

e. Continue the titration until the blue colour disappears and green colour appears. This is the endpoint of the titration.

f. Take the burette reading and then repeat the titration 2 more times to get concordant readings.

g. Perform blank determination.

9. Observations

I) Standardization of 0.02 M $KMnO_4$ solution

a) Solution in burette: 0.1 M sodium thiosulphate solution.

b) Contents of the conical flask: 20.0 mL of prepared $KMnO_4$ solution + 2 g potassium iodide + 10 mL 1 M 4 sulfuric acid solution.

c) End Point 1: Yellow-green colour, then add 3 mL starch solution as an indicator.

d) End Point 2: Blue colour disappears to green colour.

Observation table I

Sr. No	Initial burette reading (mL)	Final burette reading (mL)	Difference (mL)	Mean burette reading (MBR) (mL)
1.				
2.				
3.				

II) Blank Titration

a) Solution in burette: 0.1 M sodium thiosulphate solution.

b) Contents of the flask: 20 mL water + 2 g potassium iodide + 10 mL 1 M sulfuric acid solution.

c) End Point 1: Yellow-green colour, then add 3 mL of starch solution as an indicator.

d) End Point 2: Blue colour disappears to green colour.

Observation table II

Sr. No	Initial burette reading (mL)	Final burette reading (mL)	Difference (mL)	Mean burette reading (MBR) (mL)
1.				
2.				
3.				

Burette Reading (V_2) = MBR of standardization of $KMnO_4$ (I) - MBR of blank titration (II)

Burette Reading (V_2) = _____ - _____ = _____ mL

Calculations

Potassium permanganate = Sodium thiosulphate

$M_1 \times V_1 = M_2 \times V_2$

$M_1 = M_2 \times V_2 / V_1$

$M_1 = 0.1 \times V_2 / 20$

$M_1 = 0.005 \times V_2$

M_1 = _____

10. Result

The molarity of the prepared $KMnO_4$ solution was found to be_____ M.

11. Conclusion

Potassium permanganate solution was prepared and standardized as per the procedure given in IP 2022.

12. Reference

a. Indian Pharmacopoeia 2022.

13. Practical Related Questions

a. What is the endpoint in $KMnO_4$ standardization?
b. Can you prepare a standard solution of potassium permanganate? give reason.
c. Why is heating essential in the preparation of the $KMnO_4$ solution?
d. Define redox titration?
e. Describe the principle involved in permanganometry.

(*Space for Answers*)

14. Assessment Scheme

Particular	Understanding the basic concept (Intellectual skill)	Activities (Motor skill)	Cleanliness & Handling (Affective domain)	Viva-voce / Answers Written	Total	Signature of teacher
Marks Obtained						
Max Marks	**02**	**05**	**01**	**02**	**10**	

Experiment No. 10
Assay of Ferrous sulphate

1. Aim
To perform the assay of ferrous sulphate as per IP-2022.

2. Practical Significance
The assay of organic and inorganic pharmaceuticals is crucial for guaranteeing the quality, safety, and effectiveness of these products. These assays are vital for quality assurance, meeting regulatory standards, detecting impurities, verifying therapeutic efficacy, conducting stability tests, optimizing processes, enhancing economic efficiency, and supporting research and development. Pharmaceutical assays ensure that products meet the high-quality and purity standards required by regulatory agencies. In this experiment, students will perform an assay of ferrous sulphate using redox titration.

3. Practical Outcomes
After completion of this practical, the students will be able to:

PrO	Practical Outcomes	Mapped CO	BTL
1	Explain the principle and reaction involved in cerimetry titration.	CO2,3	2
2	Prepare 0.1 M ceric ammonium nitrate solution.	CO2,3	3
3	Perform the standardization and calculate the exact molarity of the prepared solution.	CO2,3	4
4	Perform the assay and calculate the percentage purity of ferrous sulfate.	CO2,3	5
5	Demonstrate working as a leader or team member.	CO2,3	5

4. Relevant Theoretical Background
Redox titrations involve the determination of the concentration of an analyte (the substance being analyzed) through a redox reaction. Redox titrations are based on oxidation-reduction (redox) reactions between the analyte and the titrant (a solution of known concentration). In these reactions, electrons are transferred from one substance to another. The substance being oxidized loses electrons (oxidation), while the substance being reduced gains electrons (reduction).

The assay of ferrous sulphate is a redox type of titration. Oxidation and reduction occur simultaneously in redox reactions. The reactant that loses electron(s) is a reducing agent and it can be converted to a higher state of oxidation.

In the assay of ferrous sulphate, Fe^{2+} (ferrous) ions are readily oxidized by ceric ammonium nitrate solution in acidic solution (H_2SO_4) into Fe^{3+} (ferric) ions. Thus, ferrous sulphate acts as a reducing agent. Official book Indian Pharmacopoeia 2022, mention the assay of ferrous sulphate by titrating against 0.1 M solution of ceric ammonium nitrate, using ferroin as an indicator. This method is also called cerimetry.

$$Ce^{4+} + \underset{\text{Ferrous}}{Fe^{2+}} \longrightarrow Ce^{3+} + \underset{\text{Ferric}}{Fe^{3+}}$$

5. Requirements

Glasswares: Burette (50 mL or 100 mL), Measuring cylinder (100 mL), Conical flask (250 mL), Dropper, Burette stand, Volumetric flask (1000 mL).

Chemicals: Ferrous sulphate, Nitric acid, Ceric ammonium nitrate, Distilled water, Sodium oxalate, Hydrochloric acid, Sulphuric acid, Ferroin solution.

Reagents

a. **1 M Nitric acid:** Dilute 62.5 mL of concentrated nitric acid to 1000 mL with water.

6. Requirements used

_____

_____

7. Precautions

a. Before filling the burette for the titration, rinse it with distilled water and then pre-rinse it with a portion of the titrant solution.
b. Remove air bubbles from the burette and adjust the reading to zero.
c. Do not pipette solutions by mouth.

8. Procedure

Step I) Preparation and standardization of 0.1 M Ceric Ammonium Nitrate solution

For the preparation of the solution, dissolve 54.82 g of Ceric Ammonium Nitrate in 1000 mL of 1 M nitric acid and filter.

Standardize the solution in the following manner

a. Weigh and transfer about 0.2 g of dried sodium oxalate, to a 250 mL conical flask.
b. Add 100 mL of water, then add 2 mL of conc. sulphuric acid mix well.
c. Add 10 mL of conc. hydrochloric acid mix well.
d. Heat the solution at 75ºC for 1 min.
e. Titrate with 0.1 M ceric ammonium nitrate solution until it becomes faintly yellow. Report the burette reading two more times and calculate the Molarity.

Step II) Assay of ferrous sulphate

a. Dissolve 2.5 g of sodium bicarbonate in a mixture of 150 mL of water and 10 mL of sulphuric acid in a volumetric flask.
b. When effervescence ceases, add accurately weighed 0.5 g of ferrous suphate sample.
c. Shake gently to dissolve and titrate against 0.1 M Ceric ammonium nitrate, using 0.1 mL of ferroin solution, until the red colour disappears.
d. Report the burette reading two more times and calculate the percentage purity.

9. Observations

Step I) Standardization of 0.1 M ceric ammonium nitrate (CAN)

a) Solution in burette: Prepared ceric ammonium nitrate solution.

b) Contents of the conical flask: 0.2 g sodium oxalate + 100 mL water + 2 mL conc. sulphuric acid + 10 mL of conc. hydrochloric acid + heat 75ºC for 1 min.

c) End Point 1: Faint yellow colour.

Observation table I

Sr. No	Initial burette reading (mL)	Final burette reading (mL)	Difference (mL)	Mean burette reading (MBR) (mL)
1.				
2.				
3.				

Calculations

Molarity of ceric ammonium nitrate (CAN)

Factor

1 molecule of CAN reacts with 1 molecule of Sodium oxalate

Mol. wt. of Sodium oxalate = 134 g

CAN (548.22 g) ≅ sodium oxalate (134 g)

Therefore, 1000 mL of 1 M CAN (548. 22 g dissolved in 1000 mL) = 134 g of sodium oxalate

So,

1 mL of 1 M solution of CAN = 0.134 g of sodium oxalate

1 mL of 0.1 M solution of CAN = 0.0134 g of sodium oxalate (factor)

$$\text{Molarity of ceric ammonium nitrate (m)} = \frac{\text{weight of Sodium Oxalate(g)} \times 0.1}{\text{Ceric Ammonium nitrate solution consumed (mL)} \times 0.0134 \text{ (factor)}}$$

m = _____

m = _____**M**

Step II) Assay of ferrous sulphate

a) Solution in burette: Prepared ceric ammonium nitrate solution.

b) Contents of the flask: 2.5 g sodium bicarbonate in mixture of 150 mL water + 10 mL sulphuric acid + 0.5 g ferrous suphate sample + 0.1 mL ferroin solution as an indicator.

c) End Point 1: Red colour disappears.

Observation table II

Sr. No	Initial burette reading (mL)	Final burette reading (mL)	Difference (mL)	Mean burette reading (MBR) (mL)
1.				
2.				
3.				

Calculations

Percentage purity of ferrous sulphate

Factor

1 mL of 0.1 M solution of Ceric Ammonium Nitrate is equivalent to 0.02780 g of $FeSO_4 .7H_2O$

Calculate % purity by following formula

$$\% \text{ Purity} = \frac{\text{Factor} \times \text{V} \times \text{m} \times 100}{\text{W} \times \text{M}}$$

[V=volume of CAN in assay (mean burette reading for assay), m=calculated molarity, W=weight of ferrous sulphate, M=known or actual molarity]

$$= \frac{0.02780 \times V \times m \times 100}{W \times M}$$

$$= \frac{2.780 \times V \times m}{0.5 \times 0.1}$$

= 55.6_____×_____

=_____ % w/w of $FeSO_4.7H_2O$

10. Result

a. The molarity of prepared Ceric Ammonium Nitrate (CAN) solution was found to be _____M.

b. The given sample of ferrous sulphate was found to contain _____% w/w of $FeSO_4.7H_2O$.

11. Conclusion

Assay of ferrous sulphate was carried out as per the procedure given in IP 2022.

12. References

a. Indian Pharmacopoeia 2022.

13. Practical Related Questions

a. Explain the principle for this assay.
b. Enlist the uses of ferrous sulphate.
c. Apart from cerimetry, describe any other method for assay of ferrous sulphate.
d. Describe oxidation-reduction titration.
e. Calculate the factor for the assay of ferrous sulphate by cerimetry.

(*Space for Answers*)

14. Assessment Scheme

Particular	Understanding the basic concept (Intellectual skill)	Activities (Motor skill)	Cleanliness & Handling (Affective domain)	Viva-voce / Answers Written	Total	Signature of teacher
Marks Obtained						
Max Marks	**02**	**05**	**01**	**02**	**10**	

Experiment No. 11
Assay of Calcium gluconate

1. Aim
To perform the assay of calcium gluconate as per IP-2022.

2. Practical Significance
The assay of organic and inorganic pharmaceuticals is crucial for guaranteeing the quality, safety, and effectiveness of these products. These assays are vital for quality assurance, meeting regulatory standards, detecting impurities, verifying therapeutic efficacy, conducting stability tests, optimizing processes, enhancing economic efficiency, and supporting research and development. Pharmaceutical assays ensure that products meet the high-quality and purity standards required by regulatory agencies. In this experiment, students will perform an assay of calcium gluconate using complexometric titration.

3. Practical Outcomes
After completion of this practical, the students will be able to:

PrO	Practical Outcomes	Mapped CO	BTL
1	Explain the principle and reaction involved in complexometric titration.	CO2,3	2
2	Prepare 0.05 M Disodium edetate (EDTA) solution.	CO2,3	3
3	Perform the standardization and calculate the exact molarity of the prepared solution.	CO2,3	4
4	Perform the assay and calculate the percentage purity of calcium gluconate.	CO2,3	5
5	Demonstrate working as a leader or team member.	CO2,3	5

4. Relevant Theoretical Background
Complexometric titration is a type of titration used to determine the concentration of metal ions in a solution. It involves the reaction between a metal ion and a complexing agent, also known as a chelating agent, to form a stable complex.

The assay of Calcium gluconate is complexometric titration involving complex formation reaction with EDTA (disodium edetate). There are three types of EDTA titration such as direct titration, back titration and replacement titration.

The estimation of calcium gluconate is an example of direct titration. Simple metal ion such as Ca^{2+} is transformed into complex ion by addition of a reagent which is known as 'ligand' (complexing agent). Metal ion accept electrons and ligand donates it. Disodium edetate is multidentate ligand which can form complex with metal ion by donating lone pair of electrons in presence of strong ammonia solution The end point is determined by addition of mordant black-II as an indicator, the colour changes from red to blue.

$$Ca^{2+} + \text{EDTA} \xrightarrow[\text{pH 10}]{\text{Ammonia Buffer}} \text{Calcium Edetate Complex}$$

5. Requirements

Glasswares: Burette (50 mL or 100 mL), Measuring cylinder (50mL, 10 mL), Conical flask (250 mL), beaker (250 mL), Dropper, Burette stand, Volumetric flask (1000 mL×1, 100 mL × 3)

Chemicals: Mordant Black II, Ammonia buffer, Magnesium sulphate, Sodium hydroxide, Disodium edetate, Granulated zinc, Bromine water, Calcium gluconate.

Reagents

a. **2 M Sodium hydroxide:** Weigh 8.2 g and dissolve in 50 mL distilled water, cool and make up to 100 mL in volumetric flask.
b. **Ammonia buffer solution:** Dissolve 7.0 g of Ammonium Chloride in 57.0 mL concentrated ammonia solution and dilute to 100 mL with distilled water.
c. **0.05 M Magnesium sulphate:** Weigh 0.6 g of anhydrous $MgSO_4$ and dissolve in 50 mL of distilled water, mix properly. Once it has completely dissolved, make up the volume to 100 mL, in a volumetric flask.

6. Requirements used

_____

_____

7. Precautions

a. Before filling the burette for the titration, rinse it with distilled water and then pre-rinse it with a portion of the titrant solution.
b. Remove air bubbles from the burette and adjust the reading to zero.
c. Do not pipette solutions by mouth.
d. Stored solution of Disodium edetate, has validity of 1 month. Always re-standardize the solution before use.

8. Procedure

Step I) Preparation and standardization of 0.05 M Disodium edetate (EDTA) solution

Dissolve 18.6 g of Disodium edetate in sufficient water to produce 1000 mL.

Standardize the solution in the following manner

a. Weigh accurately about 0.4 g of granulated zinc, dissolve by gentle warming in 12 mL of dilute HCl solution and 0.05 mL of bromine water.
b. Boil to remove excess bromine, cool and add sufficient water to produce 200 mL.
c. Pipette out 20 mL of this resulting solution into a flask and neutralize with 2 M sodium hydroxide solution.
d. Dilute to about 150 mL with water, add sufficient ammonia buffer pH 10 to dissolve the precipitate and add 5 mL in excess.
e. Add 50 mg of Mordant black II (Erichrome black T) to the mixture and titrate against the disodium edetate solution until the solution turns green.
f. Perform blank titration.

[*The disodium salt of ethylene diamine tetraacetic acid is preferred due to purity reasons. Bromine solution is added to ensure oxidation of trace impurity of iron (II) of iron (III), Which forms a much less stable edetate complex than iron (II)]*

Step II) Assay of Calcium gluconate

a. Weigh calcium gluconate about 0.5 g and dissolve in 50 mL of warm water.
b. Cool, and add 5.0 mL of 0.05 M magnesium sulphate and 10 mL of strong ammonia solution
c. Titrate against 0.05 M disodium edetate using Mordant black -II mixture as an indicator.
d. End point - colour changes from red to blue.
e. Perform blank titration (without calcium gluconate).
f. Repeat the assay two more times for concordat burette reading.

9. Observations

Step I) Standardization of 0.05 M Disodium edetate (EDTA) solution

a. Solution in burette: prepared disodium edetate solution.

b. Contents in beaker: 0.4 g granulated zinc + 12 mL dilute HCl solution + 0.05 mL of bromine water boil and cool + add sufficient water to produce 200 mL total solution.

c. Contents of the flask: 20 mL of above solution + neutralize with 2 M sodium hydroxide solution + dilute to 150 mL using sufficient water + add sufficient ammonia buffer pH 10 to dissolve the precipitate + 50 mg of Mordant black II as an indicator

d. End Point: Green colour.

Observation table I

Sr. No	Initial burette reading (mL)	Final burette reading (mL)	Difference (mL)	Mean burette reading (MBR) (mL)
1.				
2.				
3.				

Step II) Blank titration

Perform the step-I **WITHOUT** granulated zinc.

Observation table II

Sr. No	Initial burette reading (mL)	Final burette reading (mL)	Difference (mL)	Mean burette reading (MBR) (mL)
1.				
2.				
3.				

Burette Reading (B.R.) = MBR for standardization of disodium edetate (step I) – MBR of blank titration (step II)

Burette reading (B.R.) = _____ - _____ = _____mL

Calculations

Factor

1 molecule of EDTA reacts with 1 molecule of Zn

EDTA (372 g) ≅ Zn (65.4 g)

1000 mL of 1 M EDTA (372 g in 1000 mL water) = 65.4 g of Zn

Therefore,1 mL of 1 M solution of EDTA = 0.0654g of Zn

1 mL of 0.05 M disodium edetate (EDTA) is equivalent to 0.00327 g of Zn.

Calculate molarity in following way

$$\text{Molarity of EDTA (m)} = \frac{\text{Weight of granulated zinc in 20 mL water (g) x 0.05}}{\text{EDTA solution consumed (B. R.) (mL)} \times 0.00327}$$

$$\text{Molarity of EDTA (m)} = \frac{0.04 \text{ x } 0.05}{\text{B. R.} \times 0.00327} = \frac{0.612}{\text{B. R.}} = \text{\_\_\_\_\_}$$

m = _____ **M**

Step III) Assay of calcium gluconate

a. Solution in burette: Prepared disodium edetate solution.

b. Contents of the flask: 0.5 g calcium gluconate + 50 mL warm water + 5.0 mL 0.05 M Magnesium sulphate + 10 mL strong ammonia solution + 50 mg of Mordant black II as an indicator.

c. End Point: Colour changes from red to blue.

Observation table III

Sr. No	Initial burette reading (mL)	Final burette reading (mL)	Difference (mL)	Mean burette reading (MBR) (mL)
1.				
2.				
3.				

Step IV) Blank Titration

Perform the step-III **WITHOUT** calcium gluconate.

Observation table IV

Sr. No	Initial burette reading (mL)	Final burette reading (mL)	Difference (mL)	Mean burette reading (MBR) (mL)
1.				
2.				
3.				

Burette Reading (V) = MBR assay of calcium gluconate (step III) – MBR of blank titration (step IV).

Burette reading (V) = _____ - _____ = _____mL

Calculations

Factor

1 mL of 0.05 M disodium edetate (EDTA) is equivalent to 0.02242 g of $C_{12}H_{12}O_{14}$ Ca.H_2O

Calculate % purity by following formula

$$\% \text{ Purity} = \frac{\text{Factor} \times \text{V} \times \text{m} \times 100}{\text{W} \times \text{M}}$$

[V=volume of EDTA in assay (mean burette reading-blank burette reading for assay), m= calculated molarity, W= weight of calcium gluconate (g), M= known or actual molarity]

$$= \frac{0.02242 \text{ x } V \text{ x } m \text{ x } 100}{0.5 \text{ x } 0.05}$$

$$= \frac{2.242 \text{ x } V \text{ x } m}{0.025}$$

= 86.68_____×_____

=_____

10. Result

a. The molarity of prepared EDTA (Disodium edetate) solution was found to be _____M.

b. The given sample of calcium gluconate was found to contain _____% w/w of $C_{12}H_{12}O_{14}$ $Ca.H_2O$.

11. Conclusion

Assay of calcium gluconate was carried out as per the procedure given in IP 2022.

12. References

a. Indian Pharmacopoeia 2022.

13. Practical Related Questions

a. Apart from EDTA give the examples of chelating agents that are used in complexometric titrations.
b. Describe principle for the assay of calcium gluconate.
c. Draw the structure of calcium gluconate.
d. Write the factor calculation for calcium gluconate assay.

(*Space for Answers*)

14. Assessment Scheme

Particular	Understanding the basic concept (Intellectual skill)	Activities (Motor skill)	Cleanliness & Handling (Affective domain)	Viva-voce / Answers Written	Total	Signature of teacher
Marks Obtained						
Max Marks	**02**	**05**	**01**	**02**	**10**	

Experiment No. 12
Assay of Sodium chloride

1. Aim
To perform the assay of Sodium chloride as per IP-2022.

2. Practical Significance
The assay of organic and inorganic pharmaceuticals is crucial for guaranteeing the quality, safety, and effectiveness of these products. These assays are vital for quality assurance, meeting regulatory standards, detecting impurities, verifying therapeutic efficacy, conducting stability tests, optimizing processes, enhancing economic efficiency, and supporting research and development. Pharmaceutical assays ensure that products meet the high-quality and purity standards required by regulatory agencies. In this experiment, students will perform an assay of sodium chloride using argentometric titration.

3. Practical Outcomes
After completion of this practical, the students will be able to:

PrO	Practical Outcomes	Mapped CO	BTL
1	Explain the principle and reaction involved in argentometric titration.	CO2,3	2
2	Prepare 0.1 M Silver nitrate and 0.1 M ammonium thiocyanate solution.	CO2,3	3
3	Perform the standardization and calculate the exact molarity of the prepared solution.	CO2,3	4
4	Perform the assay and calculate the percentage purity of sodium chloride.	CO2,3	5
5	Demonstrate working as a leader or team member.	CO2,3	5

4. Relevant Theoretical Background

This assay is based on argentometric (precipitation) titration. In argentometric titration, silver ions react with thiocyanate ions. Modified Volhard's method is a type of back titration. In this method, excess silver nitrate is used to react with chloride-containing compounds (such as sodium chloride) in the presence of nitrobenzene or dibutyl phthalate.

After the reaction, the excess silver nitrate is titrated against an ammonium thiocyanate solution in the presence of nitric acid and ferric alum as indicators. Ammonium thiocyanate is added to the reaction mixture until a reddish-yellow color appears. This color results from the reaction of ammonium thiocyanate with the ferric alum indicator, forming a ferric thiocyanate complex.

In Volhard's method, a specific quantity of silver nitrate is added, whereas in modified Volhard's method, excess silver nitrate is added to sodium chloride, and then the unreacted silver nitrate is back-titrated.

1. $NaCl + AgNO_3 \longrightarrow AgCl\downarrow + NaNO_3 + AgNO_3$

Silver nitrate (excess) — Silver chloride — Silver nitrate (unreacted)

2. $AgNO_3 + NH_4SCN \xrightarrow{HNO_3} AgSCN + NH_4NO_3$

Silver nitrate (unreacted) — Ammonium thiocyanate

3. $3NH_4SCN + Fe^{+3} \longrightarrow Fe(SCN)_3$

Ferric ions present in indicator — Ferric thiocyanate (red-brown colour)

5. Requirements

a) Glasswares: Burette (50 mL or 100 mL), Measuring cylinder (50 mL, 10 mL), Conical flask (250 mL), Iodine flask (250 mL) Dropper, Burette stand, Volumetric flask (1000 mL × 2).

b) Chemicals: Silver nitrate, Sodium chloride, Acetic acid, Methanol, Eosin solution, Nitric acid, Dibutyl phthalate or Nitrobenzene, Ferric ammonium sulphate Indicator (Ferric alum), Ammonium thiocyanate.

c) Reagent

2 M nitric acid: Dilute 125 mL of concentrated nitric acid in sufficient water by cooling the solution, then make up the volume to 1000 mL with distilled water in a conical flask.

6. Requirements used

_____

_____

7. Precautions

a. Before filling the burette for the titration, rinse it with distilled water and then pre-rinse it with a portion of the titrant solution.
b. Remove air bubbles from the burette and adjust the reading to zero.
c. Do not pipette solutions by mouth.
d. Silver nitrate solution shall be protected from light on storage.

8. Procedure

Step I) Preparation and standardization of 0.1 M Silver nitrate

Dissolve 1.7 g of silver nitrate in sufficient quantity of distilled water (25 mL), make up 100 mL in a volumetric flask with distilled water. Protect the solution from light.

Standardize the solution in the following way.

a. Weigh 0.1 g of AR grade dried sodium chloride, transfer to conical flask and dissolve in 5 mL water.
b. Then, add 5 mL acetic acid and 50 mL methanol and 0.15 mL eosin solution. Stir the solution and titrate against 0.1 M silver nitrate solution until pink colour appears.
c. Repeat titration 2 more times.

Step II) Preparation of 0.1 M ammonium thiocyanate

Dissolve AR grade 7.6 g of NH_4SCN in sufficient water (250 mL), then make up 1000 mL volume with distilled water in a volumetric flask.

Step III) Assay of Sodium chloride

a. Weigh accurately about 0.1 g of NaCl and dissolve in 50 mL of water in a stoppered flask.
b. Add in excess i.e., about 100 mL of 0.1 M silver nitrate, 5 mL of 2 M nitric acid and 2 mL of dibutyl phthalate or 2.5 mL nitrobenzene.
c. Shake well, and then titrate the mixture against 0.1 M ammonium thiocyanate using 2 mL of ferric ammonium sulphate (ferric alum) solution as an indicator, until the colour becomes reddish yellow.
d. Perform blank titration. Repeat the process two more times.

9. Observations

Step I) Standardization of 0.1 M Silver nitrate

a. Solution in burette: Prepared silver nitrate solution.

b. Contents of the flask: 0.1 g dried sodium chloride AR + 5 mL water + 5 mL acetic acid + 50 mL methanol + 0.15 mL eosin solution as an indicator.

c. End Point: Pink colour appears.

Observation table I

Sr. No	Initial burette reading (mL)	Final burette reading (mL)	Difference (mL)	Mean burette reading (MBR) (mL)
1.				
2.				
3.				

Calculations

Factor

1 molecule of $AgNO_3$ reacts with 1 molecule of NaCl

$AgNO_3$ (169.89 g) ≅ NaCl (58.44 g)

1000 mL 1 M $AgNO_3$ Solution (169.89 g of $AgNO_3$ in 1000 mL water) = 58.44 g of NaCl

Therefore, **1 mL 0.1 M $AgNO_3$ Solution= 0.005844 g of NaCl**

Calculate molarity in following way

$$\text{Molarity of Silver nitrate (m)} = \frac{\text{Weight of NaCl (g) x 0.1}}{\text{Silver nitrate solution consumed (mL) x 0.005844(i. e. factor)}}$$

$$\text{Molarity (m)} = \frac{0.1 \text{ x } 0.1}{\text{B. R.} \times\ 0.005845} = \frac{1.711}{\text{B. R.}} = \_\_\_\_\_$$

m = _____**M**

Step II) Assay of sodium chloride (Back titration)

a. Solution in burette: 0.1 M ammonium thiocyanate solution.

b. Contents of the flask: 0.1 g NaCl + 50 mL water + 100 mL of 0.1 M silver nitrate + 5 mL of 2 M nitric acid + 2 mL of dibutyl phthalate or 2.5 mL nitrobenzene + 2 mL of ferric ammonium sulphate (ferric alum) solution as an indicator.

c. End Point: Colour becomes reddish yellow.

Observation table II

Sr. No	Initial burette reading (mL)	Final burette reading (mL)	Difference (mL)	Mean burette reading (MBR) (mL)
1.				
2.				
3.				

Step III) Blank titration for the assay of sodium chloride

Perform the step-II **WITHOUT** sodium chloride.

Observation table IV

Sr. No	Initial burette reading (mL)	Final burette reading (mL)	Difference (mL)	Mean burette reading (MBR) (mL)
1.				
2.				
3.				

Burette Reading (V) = MBR of blank titration (step III) - MBR for assay of sodium chloride (step II)

Burette reading (V) = _____ - _____ = _____mL.

Calculations

Factor

1mL of 0.1 M silver nitrate is equivalent to 0.005844 g of NaCl.

Calculate % purity by following formula

$$\% \text{ Purity} = \frac{\text{Factor} \times \text{V} \times \text{m} \times 100}{\text{W} \times \text{M}}$$

[V=volume of silver nitrate consumed in the assay (blank titration reading-back reading), m=calculated molarity of silver nitrate, W=weight of NaCl, M=known or actual molarity]

$$= \frac{0.005844 \text{ x } V \text{ x } m \text{ x } 100}{0.1 \text{ x } 0.1}$$

$$= 58.44 \text{ x } V \text{ x } m$$

$$= 58.44 \_\_\_\_\_ \times \_\_\_\_\_$$

$$= \_\_\_\_\_$$

10. Result

a. The molarity of prepared silver nitrate solution was found to be _____M.
b. The given sample of sodium chloride was found to contain _____ % w/w of NaCl.

11. Conclusion

Assay of sodium chloride was carried out as per the procedure given in IP 2022.

12. Reference

a. Indian Pharmacopoeia 2022.

13. Practical Related Questions

a. Describe back titration.
b. Explain the principle and reaction involved in the assay of sodium chloride.
c. Name other methods for finding % of NaCl in the given sample.
d. State difference between Volhard's method and modified Volhard's method.
e. Describe the role of nitrobenzene and acetic acid in this experiment.

(*Space for Answers*)

14. Assessment Scheme

Particular	Understanding the basic concept (Intellectual skill)	Activities (Motor skill)	Cleanliness & Handling (Affective domain)	Viva-voce / Answers Written	Total	Signature of teacher
Marks Obtained						
Max Marks	**02**	**05**	**01**	**02**	**10**	

Experiment No. 13
Assay of Ascorbic acid

1. Aim
To perform the assay of Ascorbic acid as per IP-2022.

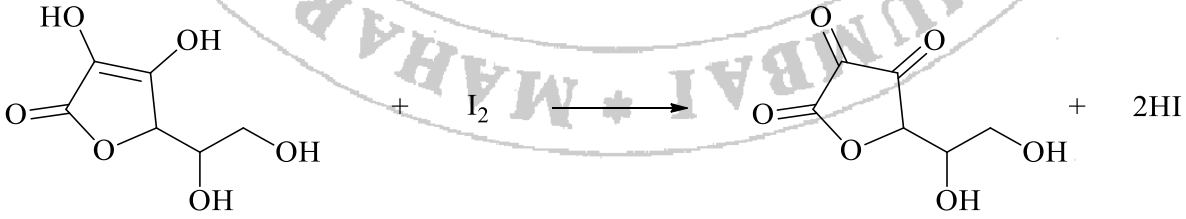
2. Practical Significance
The assay of organic and inorganic pharmaceuticals is crucial for guaranteeing the quality, safety, and effectiveness of these products. These assays are vital for quality assurance, meeting regulatory standards, detecting impurities, verifying therapeutic efficacy, conducting stability tests, optimizing processes, enhancing economic efficiency, and supporting research and development. Pharmaceutical assays ensure that products meet the high-quality and purity standards required by regulatory agencies. In this experiment, students will perform an assay of ascorbic acid using redox titration.

3. Practical Outcomes
After completion of this practical, the students will be able to:

PrO	Practical Outcomes	Mapped CO	BTL
1	Explain the principle and reaction involved in redox titration.	CO2,3	2
2	Prepare 0.05 M iodine solution.	CO2,3	3
3	Perform the standardization and calculate the exact molarity of the prepared solution.	CO2,3	4
4	Perform the assay and calculate the percentage purity of ascorbic acid.	CO2,3	5
5	Demonstrate working as a leader or team member.	CO2,3	5

4. Relevant Theoretical Background
Redox titration is a type of titration used to determine the concentration of a substance (analyte) by reacting it with a strong oxidizing or reducing agent. The reaction involves the transfer of electrons between the analyte and the titrant, causing a colour change or other detectable signal.

Assay of ascorbic acid is a type of redox titration, in this iodine oxidizes the ascorbic acid (reducing agent) into dehydroascorbic acid, at the endpoint iodine react with starch mucilage to give blue colour.



Ascorbic acid Dehydro ascorbic acid

5. Requirements
Glasswares: Burette (50 mL or 100 mL), Measuring cylinder (100 mL), Conical flask (250 mL× 2), Dropper, Burette stand, Volumetric flask (1000mL × 4).

Chemicals: Sodium hydroxide, Sulfuric acid, Hydrochloric acid, Starch, Methyl orange solution, Sodium carbonate, Iodine, Potassium iodide, arsenic trioxide.

Reagents

a. **1 M sodium hydroxide:** Weigh 42 g and dissolve in 500 mL distilled water, cool and make up volume to 1000 mL in a volumetric flask.
b. **Dilute hydrochloric acid:** Dilute approximately 100 mL of concentrated HCl to 1000 mL water.
c. **1M sulfuric acid:** Add 54 mL of concentrated sulphuric acid carefully to 100 mL of water, then make up 1000 mL with water in a volumetric flask.
d. **Starch Mucilage:** Triturate 0.5 g of starch or soluble starch with 5 mL of water and add sufficient water to produce about 100 mL, stirring continuously. Boil for a few minutes, cool and filter. (It must be freshly prepared.)

6. Requirements used

_____

_____

7. Precautions

a. Before filling the burette for the titration, rinse it with distilled water and then pre-rinse it with a portion of the titrant solution.
b. Remove air bubbles from the burette and adjust the reading to zero.
c. Arsenic is a very poisonous element. The pinpoint amount can be lethal.
d. For storing iodine solution use amber colour bottles, to protect from light.

8. Procedure

Step I) Preparation and standardization of 0.05 M iodine solution

Dissolve approximately 14 g of iodine in a solution of 36 g of potassium iodide in 100 mL of water, add three drops of hydrochloric acid and dilute up to 1000 mL in a volumetric flask with distilled water. Protect the solution from light.

Standardize the solution in the following way.

a. In 250 mL conical flask transfer accurately weighed 0.15 g dried arsenic trioxide, dissolved in 20 mL of 1 M sodium hydroxide (by warming if necessary).
b. Add 40 mL of water, add 0.1 mL methyl orange solution as an indicator, and then add dropwise dilute hydrochloric acid until the yellow colour is changed to pink.
c. Add 2 g of sodium carbonate, add 50 mL of water and add 3 mL of starch solution. Titrate with the iodine solution until a permanent blue colour is produced.
d. Repeat titration two more times.

Step II) Assay of Ascorbic acid

a. Weigh accurately about 0.1 g of sample and dissolve in a mixture of 100 mL of freshly boiled and cooled water and 25 mL of 1 M sulphuric acid.
b. Shake well and then, immediately titrate with 0.05 M iodine, using starch solution as indicator until a persistent blue-violet colour is obtained.
c. Repeat the process two more times.

9. Observations

Step I) Standardization of 0.05 M iodine solution

a. Solution in burette: Prepared iodine solution.

b. Contents of the flask: 0.15 g dried arsenic trioxide + 20 mL 1 M sodium hydroxide warm if necessary + 40 mL of water + 0.1 mL methyl orange solution + dropwise dil. HCl until the yellow colour is changed to pink + 2 g sodium carbonate + 50 mL of water + 3 mL of starch solution as an indicator.

c. End Point: Blue colour.

Observation table I

Sr. No	Initial burette reading (mL)	Final burette reading (mL)	Difference (mL)	Mean burette reading (MBR) (mL)
1.				
2.				
3.				

Calculations

Factor

1 molecule of Iodine ≅ 1/2 molecule of AS_2O_3

Iodine (130 g) ≅ AS_2O_3 (197.84/2 = 98.92 g)

1000 mL 1 M I_2 Solution (130 g of Iodine in 1000 mL water) = 98.92 g of AS_2O_3

Therefore, 1 mL 0.1 M I Solution = 0.009892 g of AS_2O_3

1 mL of 0.05 M iodine is equivalent to 0.004946 g of AS_2O_3

Calculate molarity in following way

$$\text{Molarity of iodine (m)} = \frac{\text{Weight of Arsenic trioxide(g) x 0.05}}{\text{Iodine solution consumed(B. R.) x 0.004946 (i. e. factor)}}$$

$$\text{Molarity (m)} = \frac{0.15 \text{ x } 0.05}{\text{B. R.} \times\ 0.004946} = \frac{1.5163}{\text{B. R.}} = \underline{\qquad\qquad}$$

m = _____**M**

Step II) Assay of ascorbic acid

a. Solution in burette: Prepared iodine solution.

b. Contents of the flask: 0.1 g of ascorbic acid + dissolve in a mixture of 100 mL of water and 25 mL of 1 M sulphuric acid + 3 mL of starch solution as an indicator.

c. End Point: Blue-violet colour.

Observation table II

Sr. No	Initial burette reading (mL)	Final burette reading (mL)	Difference (mL)	Mean burette reading (MBR) (mL)
1.				
2.				
3.				

Calculations

Factor

1 mL of 0.05 M iodine is equivalent to 0.008806 g of $C_6H_3O_6$

Calculate % purity by following formula

$$\% \text{ Purity} = \frac{\text{Factor} \times \text{V} \times \text{m} \times 100}{\text{W} \times \text{M}}$$

[V=volume of Iodine solution consumed (B. R.), m=calculated molarity, W=weight of Ascorbic acid, M=known or actual molarity]

$$= \frac{0.008806 \text{ x } V \text{ x } m \text{ x } 100}{0.1 \text{ x } 0.05}$$

$$= 176.12 \text{ x } V \text{ x } m$$

= 176.12_____×_____

=_____

10. Result

a. The molarity of prepared Iodine solution was found to be _____M.
b. The given sample of ascorbic acid was found to contain _____ % w/w of $C_6H_3O_6$.

11. Conclusion

Assay of ascorbic acid was carried out as per the procedure given in IP 2022.

12. References

a. Indian Pharmacopoeia 2022.

13. Practical Related Questions

a. Write the uses of ascorbic acid.
b. Describe the role of NaOH in the solution of arsenic trioxide.
c. Describe the principle (with chemical equation) for the assay of ascorbic acid.

d. Calculate the factor for the assay of ascorbic acid.

(*Space for Answers*)

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14. Assessment Scheme

Particular	Understanding the basic concept (Intellectual skill)	Activities (Motor skill)	Cleanliness & Handling (Affective domain)	Viva-voce / Answers Written	Total	Signature of teacher
Marks Obtained						
Max Marks	**02**	**05**	**01**	**02**	**10**	

Experiment No. 14
Assay of Ibuprofen

1. Aim
To perform the assay of Ibuprofen as per IP-2022.

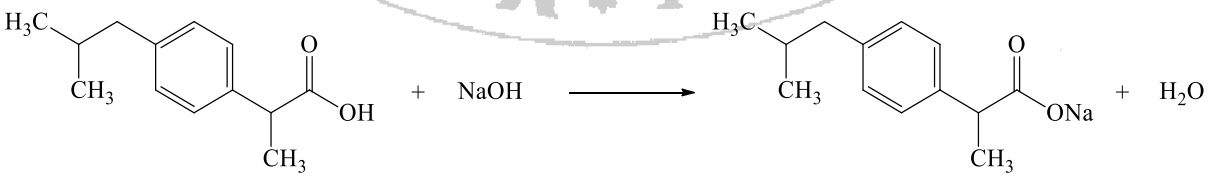
2. Practical Significance
The assay of organic and inorganic pharmaceuticals is crucial for guaranteeing the quality, safety, and effectiveness of these products. These assays are vital for quality assurance, meeting regulatory standards, detecting impurities, verifying therapeutic efficacy, conducting stability tests, optimizing processes, enhancing economic efficiency, and supporting research and development. Pharmaceutical assays ensure that products meet the high-quality and purity standards required by regulatory agencies. In this experiment, students will perform an assay of ibuprofen using alkalimetry titration.

3. Practical Outcomes
After completion of this practical, the students will be able to:

PrO	Practical Outcomes	Mapped CO	BTL
1	Explain the principle and reaction involved in alkalimetry titration.	CO2,3	2
2	Prepare 0.1 M NaOH solution.	CO2,3	3
3	Perform the standardization and calculate the exact molarity of the prepared solution.	CO2,3	4
4	Perform the assay and calculate the percentage purity of Ibuprofen.	CO2,3	5
5	Demonstrate working as a leader or team member.	CO2,3	5

4. Relevant Theoretical Background
Alkalimetry is a type of titration used to determine the concentration of an alkaline substance (base) in a solution. It involves reacting the acid with a strong base, such as sodium hydroxide, until the acid is completely neutralized.

Assay of ibuprofen is a type of alkalimetry titration (Weak acid-strong base neutralization reaction). Ibuprofen is a weak acid with a carboxylic acid group (−COOH) in its structure. In the presence of sodium hydroxide, the carboxylic acid group of ibuprofen reacts with the base to form a water-soluble salt (sodium ibuprofenate) and water.



Ibuprofen Sodium salt of ibuprofen

5. Requirements
Glasswares: Burette (50 mL or 100 mL), Measuring cylinder (50 mL, 10 mL), Conical flask (250 mL), beaker (250 mL), Dropper, Burette stand, Volumetric flask (1000 mL).

Chemicals: Phenolphthalein, Sodium hydroxide, Ibuprofen, Potassium hydrogen phthalate, Ethanol.

Reagents

a. **Phenolphthalein solution:** Dissolve 1 g of phenolphthalein in 100 mL ethanol.

6. Requirements used

_____

_____

7. Precautions

a. Before filling the burette for the titration, rinse it with distilled water and then pre-rinse it with a portion of the titrant solution.
b. Remove air bubbles from the burette and adjust the reading to zero.

8. Procedure

Step I) Preparation and standardization of 0.1 M NaOH

Dissolve 4.2 g of NaOH in sufficient carbon dioxide free water (200 mL), make up the volume to 1000 mL with distilled water in volumetric flask.

Standardize the solution in the following way.

a. Weigh accurately 1 g of pure and dried potassium hydrogen phthalate by the method of difference. Transfer in a dry conical flask.
b. Dissolve in 15 mL of carbon-dioxide free water.
c. Add one drop of Phenolphthalein indicator.
d. Fill a clean burette with 0.1 M NaOH solution upto zero mark.
e. Place the flask below the burette, add slowly 0.1 M NaOH solution dropwise until the solution in the flask is faintly pink. Take burette reading.
f. Repeat this process for 2 more times.

Step II) Assay of Ibuprofen

a. Weigh 0.4 g of ibuprofen sample and then, dissolve in 100 mL of ethanol (95 %).
b. Titrate with 0.1 M sodium hydroxide using 0.2 mL of phenolphthalein solution as an indicator.
c. Colour changes to faint pink at the end of titration.
d. Carry out a blank titration.
e. Repeat the process 2 more times.

9. Observations

Step I) Standardization of 0.1 M NaOH solution

a. Solution in burette: Prepared NaOH solution.

b. Contents of the flask: 1 g KHP + 15 mL water + drop of phenolphthalein solution as an indicator.

c. End Point: Colourless to faint pink colour.

Observation table I

Sr. No	Initial burette reading (mL)	Final burette reading (mL)	Difference (mL)	Mean burette reading (MBR) (mL)
1.				
2.				
3.				

Calculations

Factor

1 mL of 0.1 M NaOH = 0.020423 g of KHP.

Calculate molarity in following way

$$\text{Molarity of NaOH (m)} = \frac{\text{Weight of KHP (g) x 0.1}}{\text{NaOH solution consumed(B. R.) x 0.020423 (i. e. factor)}}$$

$$\text{Molarity (m)} = \frac{1 \text{ x } 0.1}{\text{B. R.} \times 0.020423} = \frac{4.896}{\text{B. R.}} = \text{\_\_\_\_\_}$$

m = _____**M**

Step II) Assay of Ibuprofen

a. Solution in burette: Prepared NaOH solution.

b. Contents of the flask: 0.4 g ibuprofen + 100 mL of ethanol + 0.2 mL phenolphthalein solution as an indicator.

c. End Point: Faint pink colour.

Observation table II

Sr. No	Initial burette reading (mL)	Final burette reading (mL)	Difference (mL)	Mean burette reading (MBR) (mL)
1.				
2.				
3.				

Calculations

Factor

1 mL of 0.1 M NaOH is equivalent to 0.02063 g of ibuprofen.

Calculate % purity by following formula

$$\% \text{ Purity} = \frac{\text{Factor} \times \text{V} \times \text{m} \times 100}{\text{W} \times \text{M}}$$

[V=volume of NaOH solution consumed-blank titration reading for assay, m=calculated molarity, W=weight of ibuprofen, M=known or actual molarity]

$$= \frac{0.02063 \text{ x } V \text{ x } m \text{ x } 100}{0.4 \text{ x } 0.1}$$

$$= 51.57 \text{ x } V \text{ x } m$$

= 51.57_____×_____

=_____% w/w of $C_{13}H_{18}O_2$.

10. Result

a. The molarity of prepared NaOH solution was found to be _____M.
b. The given sample of ibuprofen was found to contain _____ % w/w of $C_{13}H_{18}O_2$.

11. Conclusion

Assay of ibuprofen was carried out as per the procedure given in IP 2022.

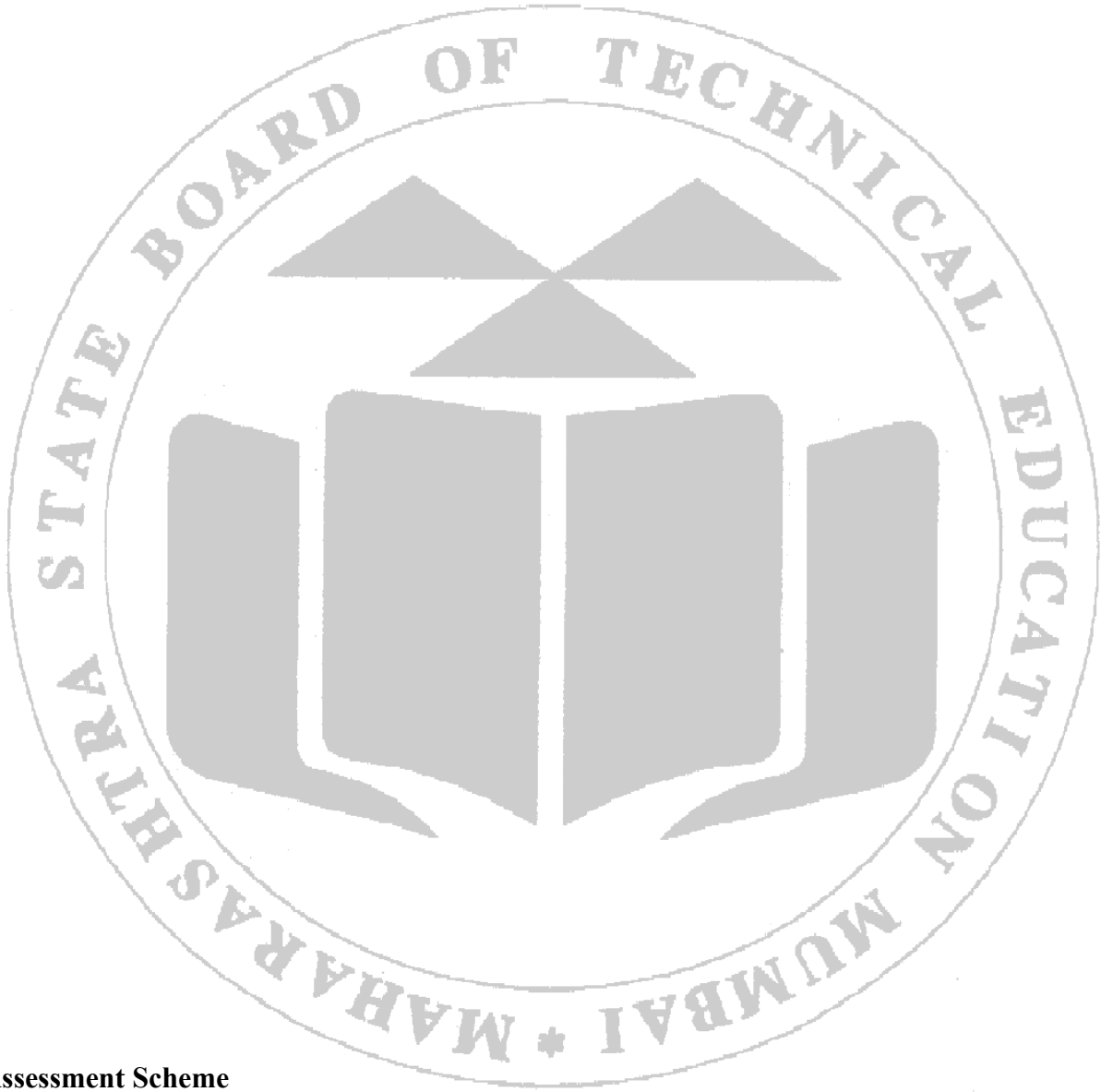
12. References

a. Indian Pharmacopoeia 2022.

13. Practical Related Questions

a. Write the uses of ibuprofen.
b. Describe the principle (with chemical equation) for the assay of ibuprofen.
c. Calculate factor for the assay of ibuprofen.
d. Define anti-inflammatory agents? Give examples.

(*Space for Answers*)



14. Assessment Scheme

Particular	Understanding the basic concept (Intellectual skill)	Activities (Motor skill)	Cleanliness & Handling (Affective domain)	Viva-voce / Answers Written	Total	Signature of teacher
Marks Obtained						
Max Marks	**02**	**05**	**01**	**02**	**10**	

Experiment No. 15
Determination of Melting Point

1. Aim
To determine the melting point of the given solid organic compound.

2. Practical Significance
The melting point serves as a crucial tool in both identifying unknown compounds and evaluating their purity. When assessing the melting point of a substance, pure crystalline organic compounds typically exhibit a sharp, well-defined melting temperature within a very narrow range, often spanning only 0.5 to 1°C. Conversely, impure or contaminated organic compounds tend to display a broader melting interval. The presence of impurities within a compound can significantly impact its melting behaviour. Even a small amount of impurity can lower the melting point, often resulting in a melting temperature lower than that of the pure substance. Additionally, impurities can broaden the melting range, making it less distinct and more variable. A narrow, sharply defined melting range suggests high purity, while a broader and lower melting range indicates the presence of impurities.

3. Practical Outcomes
After completion of this practical, the students will be able to:

PrO	Practical Outcomes	Mapped CO	BTL
1	Explain the effect of impurities on the melting point of a compound	CO3	2
2	Handle the Thiele's tube and thermometer.	CO3	3
3	Recognize the compound by determining melting point.	CO3	4
4	Demonstrate working as a leader or team member.	CO3	5

4. Relevant Theoretical Background
Melting Point: The melting point of a substance is the temperature at which solid –state changes to a liquid state. At the melting point, the solid and liquid states exist in equilibrium. It is often used to identify organic and inorganic crystalline compounds and to determine their purity. A definite and sharp melting point is an identity for pure organic compounds. Therefore, the melting point is a valuable criterion for the identification of an organic compound.

Note: Silicon oil is the safest and the most satisfactory liquid with high stability & heat resistant. Liquid paraffin is used in the determination of melting point because the boiling point of liquid paraffin is more than 370^0C.Due to its higher boiling point and low specific heat, non-corrosive nature, liquid paraffin easily reaches the desired temperature (220°C) without boiling.

5. Requirements
Thiele's tube, Thermometer, Capillary tube, Thread/rubber ring, Liquid paraffin.

6. Requirements used

_____

_____

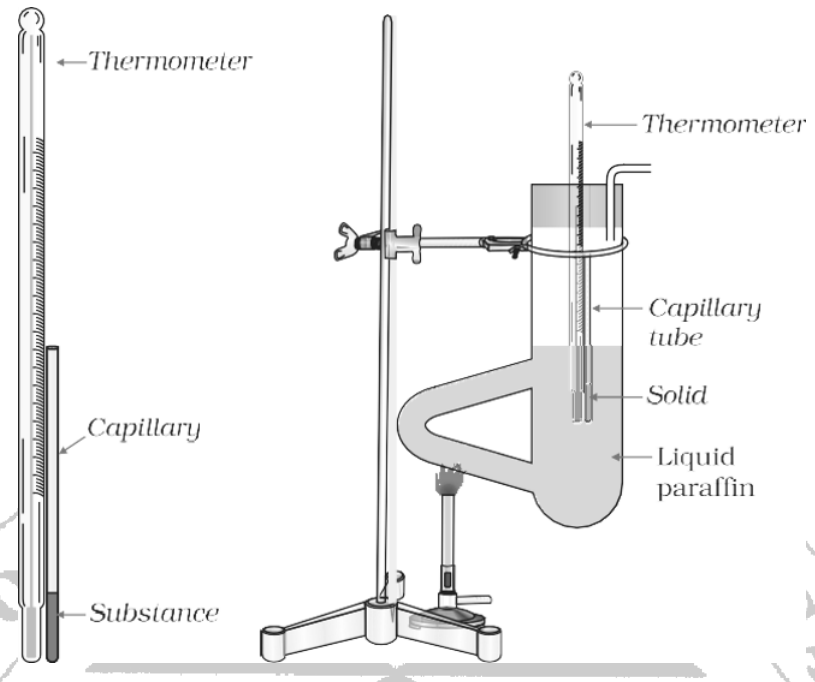


Fig 15.1: Metling Point Apparatus

7. Procedure

a. Take a capillary tube of approximately 8 cm in length. Seal its one open end by heating it in flame for a while.
b. Fill the powdered organic compound into a sealed capillary tube.
c. Tie the filled capillary tube to the thermometer near its bulb with the help of a thread/ rubber ring.
d. Take a Thiele's tube and fill it with 50 to 60 mL Liquid paraffin.
e. Dip the thermometer along with the capillary tube in liquid paraffin in such a way that the thermometer bulb and the filled portion of the capillary are completely dipped in the liquid paraffin and the open end of the capillary remains in the air. The thermometer and the capillary tube should not touch the sides of Thiele's tube.
f. Heat the side arm of the Thiele's tube with the help of a gas burner.
g. Note the temperature when the solid starts melting. This temperature is the melting point of the solid organic compound.
h. Repeat the procedure thrice and record the observations.

8. Precautions

a. Keep the capillary tube and the thermometer at a similar level.
b. Tightly pack the powder into the capillary tube without any air gaps.
c. Control the rate of heating to ensure that the sample melts uniformly and doesn't decompose. A typical heating rate is 1-2 ^{0}C / minute.
d. Use proper lighting and magnification to observe the sample during melting. Note the temperature range over which melting occurs.
e. After the melting point determination, allow the apparatus to cool before starting a new determination to prevent cross-contamination between samples.

9. Observations

Melting Point Temperature

Sr. No.	Temperature in ^{o}C
1	
2	
3	

10. Result

a. The Melting point of a given sample of organic compound was found to be_____.

b. The Melting point of a given sample of organic compound as per the official book is _____.

11. Conclusion

a. The melting point of a given sample of organic compound was recorded.

b. If the melting point of a given organic compound is not same as the value stated in official book/literature, it may be due to presence of impurities in the given organic compound. Hence the given sample is _____ (Pure/ Impure).

12. Reference

a. Vogel's Textbook of Practical Organic Chemistry, 5th Edition, By Brian S. Furniss, Antony J. Hannaford, Peter W.G. Smith, Austin R. Tatchell.

13. Practical Related Questions

a. Define Melting point.

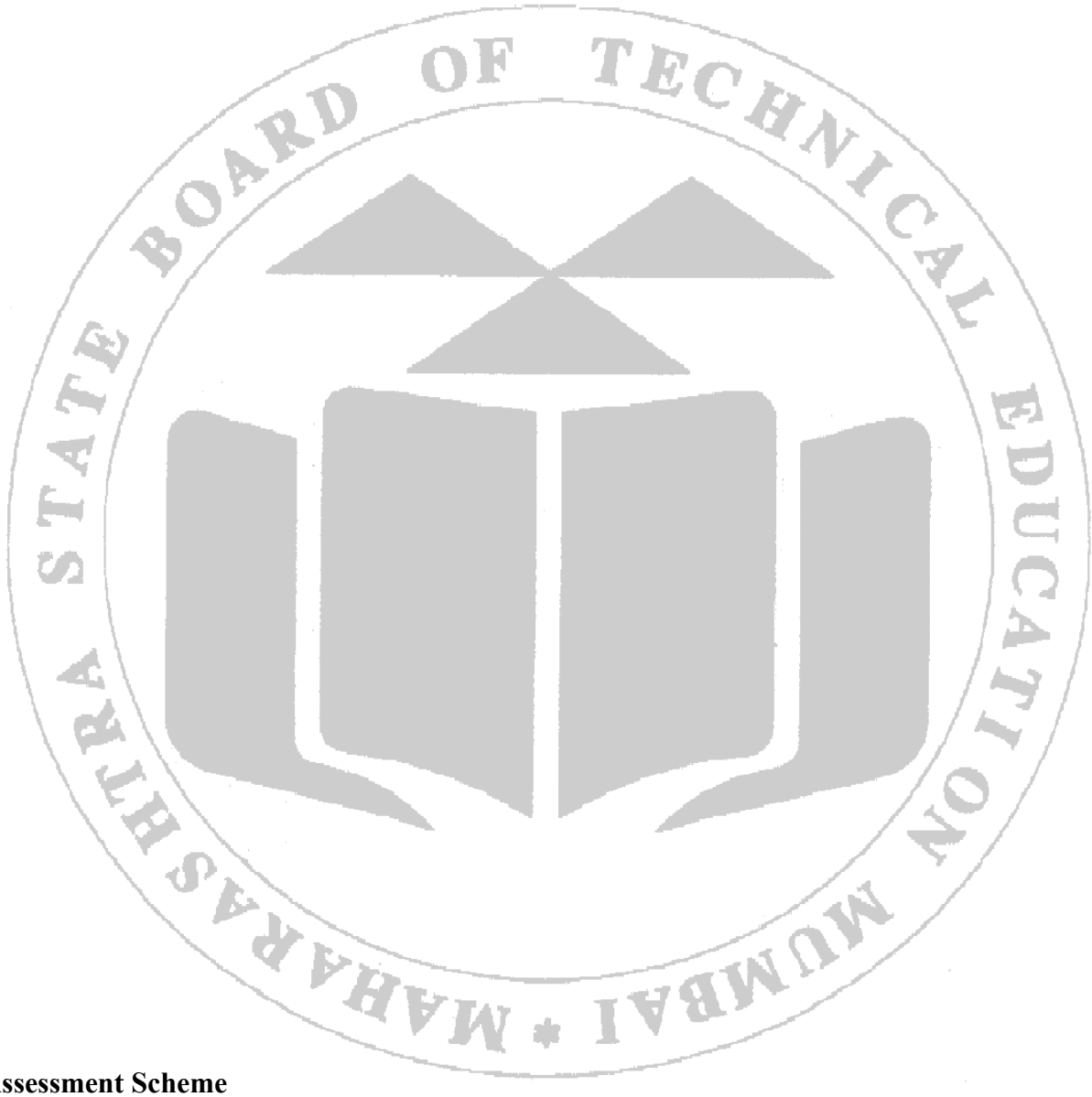
b. Why do pure solids possess sharp melting point?

c. What is the effect of impurities on the melting point of a solid?

d. Why is liquid paraffin used to determine melting point?

e. What is the melting point range of organic compound?

(*Space for Answers*)



14. Assessment Scheme

Particular	Understanding the basic concept (Intellectual skill)	Activities (Motor skill)	Cleanliness & Handling (Affective domain)	Viva-voce / Answers Written	Total	Signature of teacher
Marks Obtained						
Max Marks	**02**	**05**	**01**	**02**	**10**	

Experiment No. 16

Determination of Boiling Point

1. Aim

To determine the boiling point of the given solid organic compound.

2. Practical Significance

Boiling point of organic compounds can provide important information regarding their physical properties and structural characteristics. The boiling point helps to identify a compound and to characterize it. The boiling point of the liquid depends upon the pressure exerted upon the liquid surface. Since atmospheric pressure is different places, therefore a liquid has different boiling points at different places. For example, water reaches the standard atmospheric pressure at 100^0C. Water can boil at a lower temperature as elevation increases.

3. Practical Outcomes

After completion of this practical, the students will be able to:

PrO	Practical Outcomes	Mapped CO	BTL
1	Explain the effect of impurities on the boiling point of a compound	CO3	2
2	Handle the Thiele's tube and thermometer.	CO3	3
3	Recognize the compound by determining boiling point.	CO3	4
4	Demonstrate working as a leader or team member.	CO3	5

4. Relevant Theoretical Background

Boiling Point: The boiling point of a liquid may be defined as the temperature at which the vapour pressure of the liquid is equal to the atmospheric pressure. Different liquids have different boiling points. The difference in the boiling points of liquids is essentially due to the difference in the intermolecular forces operating between the molecules of the liquid. The boiling point of a liquid increases if non-volatile impurities are present in it.

Note: For practical applications, liquid paraffin serves as an ideal medium for determining melting /boiling points due to its exceptionally high boiling point exceeding 370°C. This characteristic allows precise temperature control within the desired range of 200-250°C without the risk of boiling.

5. Requirements

Thiele's tube, Thermometer, Capillary tube, Ignition tube, Thread/rubber ring, Liquid paraffin or Conc. Sulfuric acid.

6. Requirements used

_____

_____

7. Procedure

a. Take a Thiele's tube and fill it with 50 to 60 mL Liquid paraffin.

b. Take 1-2 drops of the given liquid in an ignition tube and tie the ignition tube to the thermometer with the thread or rubber ring. Note that the lower end of the ignition tube and the thermometer bulb are at the same level.
c. Take a capillary tube of approximately 8 cm in length. Seal its one open end by heating it in flame for a while.
d. Place the capillary tube with its open end dipped in the liquid present in the ignition tube.
e. Heat the side arm of the Thiele's tube with the help of a gas burner.
f. Observe the escape of bubbles at the lower end of the capillary dipped in the liquid organic compound. Note the temperature at which bubbles start coming rapidly and continuously. This temperature is the boiling point of the liquid.
g. Repeat the procedure thrice and record the observations.

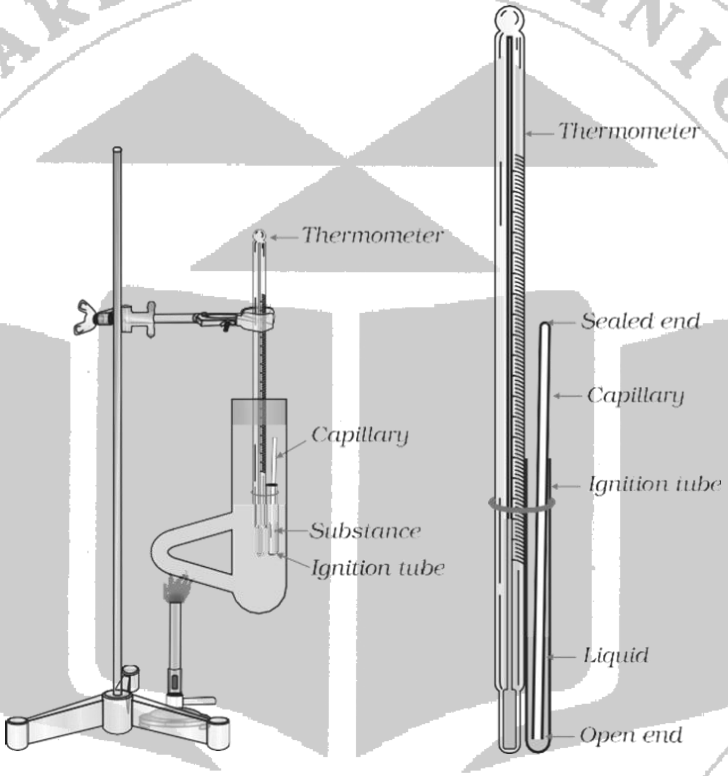


Fig 16.1: Boiling point assembly

8. Precautions

a. Control the rate of heating to ensure that the sample boils uniformly and doesn't decompose. A typical heating rate is 1-2 ^{0}C / minute.
b. Keep the ignition tube and the thermometer at a similar level.
c. Use proper lighting and magnification to observe the sample during bubbles start coming rapidly. Note the temperature range over which boiling occurs.
d. After the boiling point determination, allow the apparatus to cool before starting a new determination to prevent cross-contamination between samples.

9. Observations

Boiling Point Temperature

Sr. No.	**1**	**2**	**3**
Temperature in ^{o}C			

10. Result

a. The Boiling point of a given sample of organic compound was found to be_____.
b. The Boiling point of a given sample of organic compound as per the official book is _____.

11. Conclusion

a. The boiling point of a given sample of organic compound was recorded.
b. If the melting point of a given organic compound is not the same as the value stated in the official book/literature, it may be due to the presence of impurities in the given organic compound. Hence the given sample is _____ (Pure/ Impure).

12. Reference

a. Vogel's Textbook of Practical Organic Chemistry, 5th Edition, By Brian S. Furniss, Antony J. Hannaford, Peter W.G. Smith, Austin R. Tatchell.

13. Practical Related Questions

a. Define Boiling point.
b. Why different liquids have different boiling points?
c. What will be the effect on boiling point if two liquids are mixed?
d. What is vapour pressure?
e. What will be the effect on boiling point if atmospheric pressure is reduced?
f. What is the application of boiling point determination?

(*Space for Answers*)

14. Assessment Scheme

Particular	Understanding the basic concept (Intellectual skill)	Activities (Motor skill)	Cleanliness & Handling (Affective domain)	Viva-voce / Answers Written	Total	Signature of teacher
Marks Obtained						
Max Marks	**02**	**05**	**01**	**02**	**10**	

Experiment No. 17
Synthesis of Benzoic acid from Benzamide

1. Aim
To prepare benzoic acid from benzamide by hydrolysis and to find out its percentage practical yield and melting point.

2. Practical Significance
Organic synthesis is the artificial construction of organic molecules from smaller molecules using chemical reactions. There are various types of organic reactions used for the synthesis of new molecules. Synthetic reactions can be around two fundamental approaches: functional group interconversion and carbon-carbon bond formation. Total synthesis is a laboratory method for constructing a complex molecule, often a natural product, through a series of chemical reactions using relatively simple molecules as starting materials.

3. Practical Outcomes
After completion of this practical, the students will be able to:

PrO	Practical Outcomes	Mapped CO	BTL
1	Explain the principle and reaction involved in the synthesis of benzoic acid.	CO4	2
2	Synthesize benzoic acid from benzamide and purify it.	CO4	5
3	Calculate the required quantity of reagents and the practical percentage yield.	CO4	3
4	Identify the synthesized benzoic acid by performing melting point and chemical tests.	CO4	4
5	Demonstrate working as a leader or team member.	CO4	5

4. Relevant Theoretical Background
Synthesis: It is a process in which a new product with a unique structural formula, molecular weight, and melting point is produced with a chemical reaction.

Purification: Purification is the process of removing impurities from the product. Purification of the product includes the application of recrystallization, washing, and drying the product in an oven at a definite temperature for a desired time.

Recrystallization: Recrystallization is the process in which the compound is dissolved in selected solvent with heating and then cooled slowly to a saturated from which pure compound is crystallised out.

Yield: It is the quantity of the product obtained in the synthesis. They are:

Theoretical Yield is the weight of the product that one should get based on the stoichiometric quantities of the reagents, assuming 100% completion of the reaction.

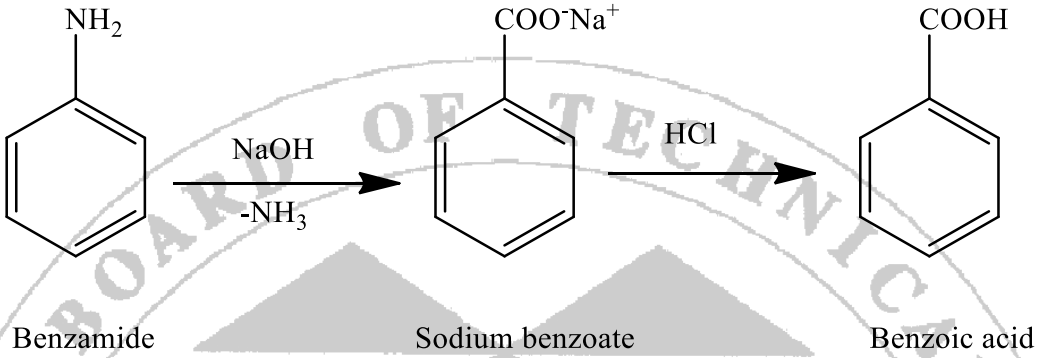
Practical Yield is the weight of the product actually obtained after purification of the product.

Percentage Yield is calculated from the formula given below:

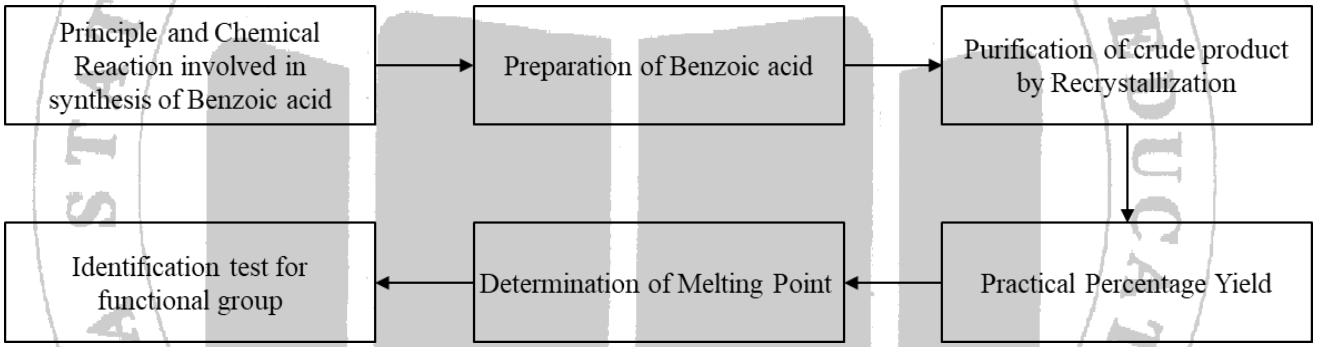
$$\% \text{ Yield} = \frac{\text{Practical Yield}}{\text{Theoretical Yield}} \times 100$$

Principle: Amides warmed with dil. NaOH gives ammonia readily together with the salt of corresponding acid**.** Benzoic acid is prepared by alkaline hydrolysis of benzamide. Hydrolysis is any chemical reaction in which one or more chemical bonds are broken with the addition of water. Carboxylic acid derivatives such as esters or amides can be hydrolyzed to carboxylic acid salt in the presence of acid or base.

Hydrolysis of Benzamide: Benzamide is hydrolyzed to sodium benzoate in the presence of sodium hydroxide and followed by acidification (Conc. HCl) for 1-3 hours to form benzoic acid.



Flow chart for Experimental Work



5. Requirements

a. Glasswares: Beaker (250 mL), Round bottom flask (250 mL), Reflux condenser, Vaccum pump with Buchner's funnel and flask, Measuring cylinder (25 mL).

b. Chemicals: Benzamide, Sodium hydroxide solution (10 %), Conc. Hydrochloric acid.

6. Requirements used

_____

_____

7. Precautions

a. Always add unglazed porcelain pieces to any solution before you begin heating it to prevent the solution from bumping.

b. A clean piece of filter paper should always be used for filtration.

8. Procedure

a. Place 5 g of benzamide and 50 mL of sodium hydroxide (3.30 g of NaOH + 50 mL) solution in a 250 mL round bottom flask fitted with a Reflux condenser.

b. Add a few pieces of unglazed porcelain into the reaction mixture.

c. Boil the mixture gently for 30 minutes.
d. Cool the solution in ice water mixture and add slowly concentrated hydrochloric acid till the mixt strongly acidic to litmus.
e. Cool the mixture in ice water for about 10 minutes and collect the product at a Buchner funnel us vacuum pump.
f. Wash with cold water and drain.
g. Recrystallize this product by dissolving in a minimum quantity of boiling water, filter the hot solution if necessary.
h. Allow to cool to room temperature. Colorless crystals of benzoic acids are obtained.
i. Collect recrystallized benzoic acid by filtration and dry it.
j. Weigh accurately the yield obtained and determine the melting point of the same.
k. Carry out identification tests for functional groups, i. e. Benzamide will show positive test for-C (Amide) group and negative test for -COOH group, while product will show test for-CONH, negative test for-COOH positive.

9. Observations

1. Amount of benzamide taken for synthesis	5 g
2. Practical yield of recrystallized product	
3. Melting point of the product	
4. Identification test for functional groups	

10. Calculations

a) Theoretical yield of the product (x g)

Molecular weight of Benzamide = 121 g
Molecular weight of Benzoic acid = 122 g
121 g of benzamide gives 122 g of benzoic acid
5 g of benzamide gives x g of benzoic acid

$$X = \frac{122 \times 5}{121} \times 100$$

Theoretical yield of Benzoic acid (x) = 5.04 g

b) Percentage practical yield

$$\% \text{ Yield} = \frac{\text{Practical Yield}}{\text{Theoretical Yield}} \times 100$$

$$\% \text{ Yield} = \frac{\qquad}{\qquad} \times 100$$

% Yield = _____%

11. Result

a. Percentage yield of benzoic acid _____%

b. Melting point of benzoic acid _____

12. Conclusion

Benzoic acid was synthesized from benzamide by hydrolysis reaction.

13. Reference

a. Vogel's Textbook of Practical Organic Chemistry, 5th Edition, By Brian S. Furniss, Antony J. Hannaford, Peter W.G. Smith, Austin R. Tatchell.

14. Practical Related Questions

a. Write the principle and reaction involved in the synthesis of benzoic acid.

b. Define the terms- Theoretical, Practical, and Percentage practical yield.

c. What is recrystallization?

d. Why are unglazed porcelain pieces added to the reaction mixture?

e. Write the uses of benzoic acid.

(*Space for Answers*)

15. Assessment Scheme

Particular	Understanding the basic concept (Intellectual skill)	Activities (Motor skill)	Cleanliness & Handling (Affective domain)	Viva-voce / Answers Written	Total	Signature of teacher
Marks Obtained						
Max Marks	**02**	**05**	**01**	**02**	**10**	

Experiment No. 18
Synthesis of Picric acid from Phenol

1. Aim
To prepare picric acid from phenol by nitration and to find out its percentage practical yield and melting point.

2. Practical Significance
Organic synthesis is the artificial construction of organic molecules from smaller molecules using chemical reactions. There are various types of organic reactions used for the synthesis of new molecules. Synthetic reactions can be around two fundamental approaches: functional group interconversion and carbon-carbon bond formation. Total synthesis is a laboratory method for constructing a complex molecule, often a natural product, through a series of chemical reactions using relatively simple molecules as starting materials.

3. Practical Outcomes
After completion of this practical, the students will be able to:

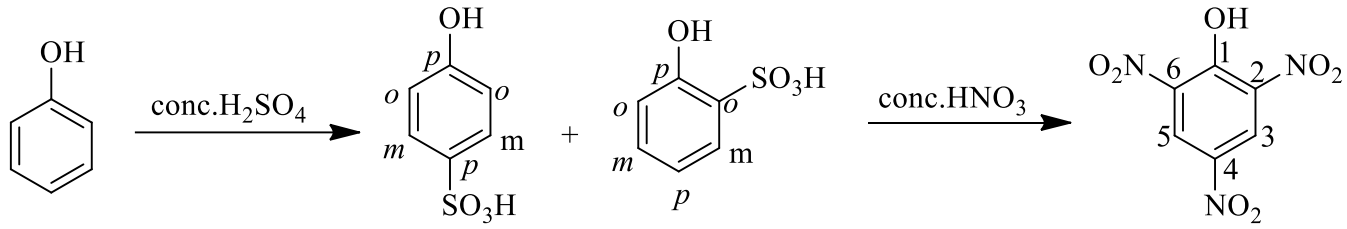
PrO	Practical Outcomes	Mapped CO	BTL
1	Explain the principle and reaction involved in the synthesis of picric acid.	CO4	2
2	Synthesize picric acid from phenol and purify it.	CO4	5
3	Calculate the required quantity of reagents and the practical percentage yield.	CO4	3
4	Identify the synthesized picric acid by performing melting point and chemical tests.	CO4	4
5	Demonstrate working as a leader or team member.	CO4	5

4. Relevant Theoretical Background
Principle:

Picric acid, also known as 2,4,6-trinitrophenol, is synthesized through a two-step process. Initially, phenol undergoes nitration in concentrated nitric acid with the assistance of concentrated sulphuric acid. This reaction is a classic example of electrophilic aromatic substitution, where a hydrogen atom in phenol is replaced by a nitro group (NO_2). The resulting nitro groups attach to the *ortho* and *para* positions of the phenol ring due to their stability.

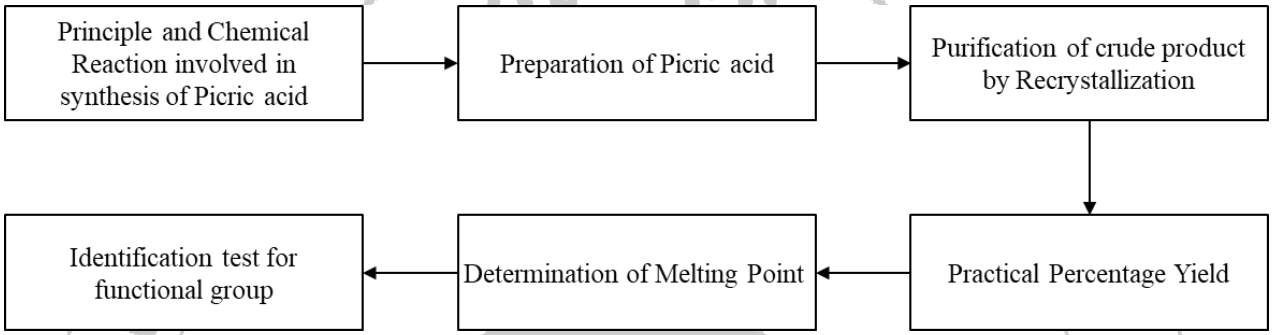
In the first step of the synthesis, phenol reacts with concentrated sulphuric acid to form mixture of *ortho* and *para* phenol sulphonic acid. Subsequently, this intermediate reacts with concentrated nitric acid, resulting in the formation of picric acid. The *ortho* and *para* positions of phenol are preferred for this reaction due to their stability, facilitating the synthesis of picric acid in high yield by the displacement of SO_3H by NO_2. The direct nitration of phenol to tri-nitro derivative in good yield is not possible since much starting material is oxidized and destroyed Picric acid finds utility in various applications, owing to its explosive and dye properties.

Chemical Reaction



Phenol | Phenol sulphonic acids | 2,4,6 trinitrophenol (Picric acid)

Flow chart for Experimental Work



5. Requirements

a. Glasswares: Beaker (250 mL), Round bottom flask (250 mL), Reflux condenser, Vaccum pump with Buchner's funnel and flask, Thiele's tube, Measuring cylinder (25 mL).

b. Chemicals: Phenol, conc. sulphuric acid, conc. Nitric acid, alcohol, water.

6. Requirements used

_____

_____

7. Precautions

a. Always add unglazed porcelain pieces to any solution before you begin heating it to prevent the solution from bumping.

b. A clean piece of filter paper should always be used for filtration.

8. Procedure

a. Weigh 4 g of phenol (or take 3.4 mL of liquefied phenol) and place in a round bottom flask.

b. Add 6.0 mL of conc. sulphuric acid and mix thoroughly. This mixture becomes warm as the reaction is exothermic.

c. Heat this RBF in a water bath for 30 minutes and cool the reaction mixture in an ice-water mixture. The phenol sulphuric acid is formed.

d. Place this RBF in a fuming cupboard and add 15 mL of concentrated nitric acid and mix it by shaking for a few seconds.

e. A vigorous reaction takes place and harmless red nitrous fumes come out from the flask.

f. After the reaction subsides, heat the flask in a boiling water bath for 2 hours with constant shaking. A heavy oily layer is formed initially and then it gets converted to a crystalline mass.

g. Add 5o mL of ice-cold water and cool the reaction mixture in ice for a few minutes.

h. Collect the crude product by vaccum filtration, wash it with cold water, and drain it completely.

i. Purify the product by recrystallization in the alcohol-water mixture (2:1).
j. Separate the crystalline material by filtration and dry it in a hot air oven.
k. Carry out identification tests for functional groups i.e., phenolic -OH and NO_2 groups. Phenols will show a positive test for phenolic -OH and a negative test for -NO_2, whereas picric acid shows both tests positive.

9. Observations

1. Amount of picric acid taken for synthesis	4 g
2. Practical yield of recrystallized product	
3. Melting point of the product	
4. Identification test for functional groups	

10. Calculations

a. Theoretical yield of the product (x g)

Molecular weight of Phenol = 94 g
Molecular weight of Picric acid = 229 g
94 g of phenol gives 229 g of picric acid
4 g of benzamide gives x g of picric acid

$$x = \frac{229 \times 4}{94}$$

Theoretical yield of Benzoic acid (x) = 9.74 g

b. Percentage practical yield

$$\% \text{ Yield} = \frac{\text{Practical Yield}}{\text{Theoretical Yield}} \times 100$$

$$\% \text{ Yield} = \text{\_\_\_\_\_} \times 100$$

% Yield = _____%

11. Result

a. Percentage yield of picric acid _____%
b. Melting point of picric acid _____.

12. Conclusion

Picric acid was synthesized from phenol by nitration reaction.

13. Reference

Vogel's Textbook of Practical Organic Chemistry, 5th Edition, By Brian S. Furniss, Antony J. Hannaford, Peter W.G. Smith, Austin R. Tatchell.

14. Practical Related Questions

a. Define electrophilic aromatic substitution reaction.
b. Write the principle and reaction involved in the synthesis of benzoic acid.
c. Phenol is acidic. Give a reason.
d. Define nitration.?
e. What care should be taken while handling conc. sulphuric acid and conc. nitric acid?

(*Space for Answers*)

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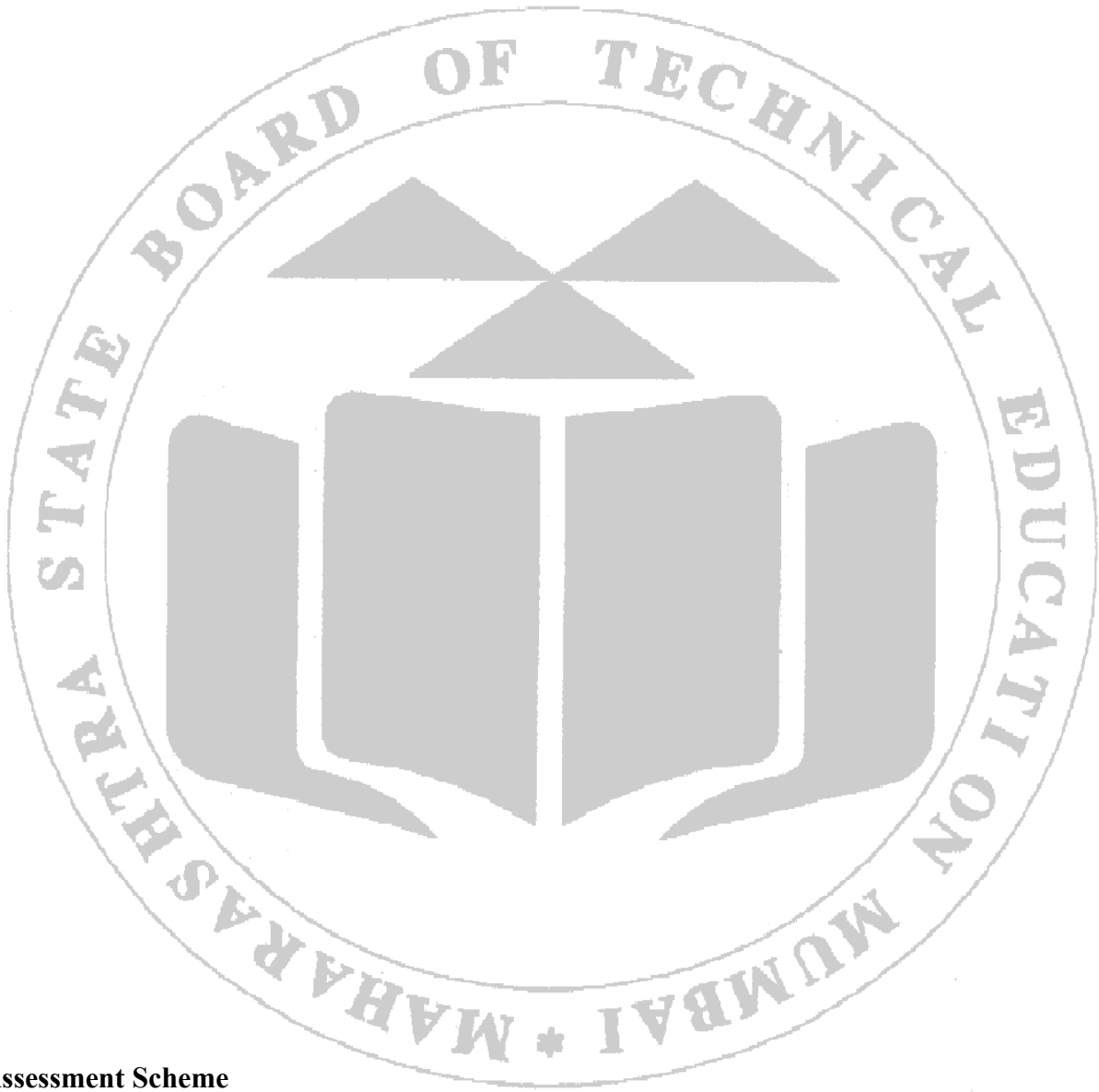


15. Assessment Scheme

Particular	Understanding the basic concept (Intellectual skill)	Activities (Motor skill)	Cleanliness & Handling (Affective domain)	Viva-voce / Answers Written	Total	Signature of teacher
Marks Obtained						
Max Marks	**02**	**05**	**01**	**02**	**10**	

Experiment No. 19
Identification test of Aspirin

1. Aim
To perform and report identification tests on the given sample of Aspirin as per IP-2022.

2. Practical Significance
The purpose of identification tests is to ensure accurate labeling of materials. Identification typically involves a combination of straightforward chemical tests and measurement of relevant physical constants. Chemical tests used for identification aim to establish the presence of specific functional groups, thereby confirming the molecular structure as accurately as possible. Inorganic substances generally rely on tests commonly used in qualitative analysis. Organic substances are identified through characteristic reactions of the functional groups present in their molecules.

3. Practical Outcomes
After completion of this practical, the students will be able to:

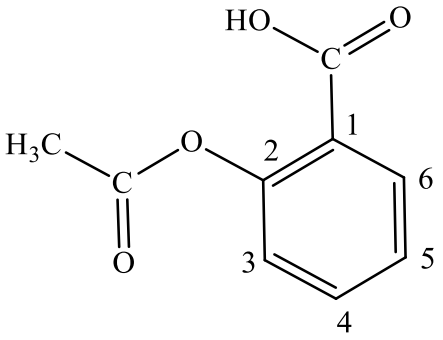
PrO	Practical Outcomes	Mapped CO	BTL
1	Describe the monograph of aspirin.	CO5	2
2	Perform identification tests on the given sample for aspirin as per IP.	CO5	5
3	Write a report on the identification test.	CO3	5
4	Follow cleanliness, safety and ethical practices.	CO5	5

4. Relevant Theoretical Background
The statement of solubility is indicated by descriptive terms and is intended to apply at 20^0C to 30^0C. The following table indicates the meaning of the terms used in statements of approximate solubility.

Descriptive Term (Statement of approximate solubility)	Approximate volume of solvent in mL /g of solute
Very soluble	Less than 1
Freely soluble	From 1 to 10
Soluble	From 10 to 30
Sparingly soluble	From 30 to 100
Slightly soluble	From 100 to 1000
Very slightly soluble	From 1000 to 10,000
Insoluble or practically insoluble	More than 10,000

Monograph of Aspirin as per IP 2022

Aspirin is acetylsalicylic acid, chemically named as 2-acetoxy benzoic acid.



Molecular formula	$C_9H_8O_4$
Molecular weight	180.2g
Standard	Caffeine contains not less than 99.5 percent and not more than 100.5 percent of $C_9H_8O_4$ calculated on the dried basis.
Organoleptic description	**Nature**- Crystals, crystalline powder **Colour**- Colourless **Odour-** Odourless **Taste**- Bitter
Solubility	Sparingly soluble-Water Freely soluble- Alcohol Soluble- Chloroform and ether
Pharmaceutical category	Non-steroidal anti-inflammatory, antirheumatic, antithrombotic
Storage	Store protected from light and moisture
Dose	As analgesic and antipyretic-300 to 600mg four to six times a day As antirheumatic-1 to 2 g, four to six times a day As antithrombotic-75 mg daily

5. Requirements

a. Glasswares: Porcelain dish, Thiele's tube, capillary, Test tubes, Glass rod

b. Chemicals: Ferric chloride, Sodium hydroxide, Alcohol (95%), Sulphuric acid, Ether, Chloroform

6. Requirements used

_____

_____

7. Precautions

a. Use clean and dry glass apparatus.

b. When sodium hydroxide comes into contact with the skin, it can trigger severe irritation and burns, potentially causing harm to both the skin and eyes. Additionally, prolonged exposure might lead to the accumulation of fluid in the lungs, a condition is known as pulmonary edema, which requires urgent medical attention.

8. Procedure

a) Organoleptic description

Observe the given drug critically for the following description.

The drug is silky white crystals, white glistering needles, or white crystalline powder, odourless as per I.P. 2022

b) Solubility test

Perform solubility tests in the different solvents. The drug is sparingly soluble in water, freely soluble in alcohol, soluble and in ether, slightly soluble in ether.

c) Identification test

Test A. Boil about 0.5 g of the drug with 10 mL of sodium hydroxide solution for 3 minutes, cool, and add 10 mL of dilute sulphuric acid, a white, crystalline precipitate is produced, it has the odour of acetic acid. Filter, dissolve the precipitate in about 2 mL of water, and add ferric chloride test solution; a deep violet colour is produced.

Test B. To the filtrate obtained in test (A) add 3 mL of alcohol (95%) and 3 mL of sulphuric acid and warm; the odour of ethyl acetate is perceptible.

Test C. Determination of the melting point: The drug melts at about 142^0C.

9. Observations

Identification test report of the sample of Caffeine

Sr. No.	Test	Observation	Inference*
1.	**Organoleptic description**		
a)	Nature		
b)	Colour		
c)	Odour		
2.	**Solubility**		
a)	Water		
b)	Alcohol		
c)	Chloroform		
d)	Ether		
3.	**Identification Test**		
a)	Test A		
b)	Test B		
c)	Test C		

*If observation is as per given Indian Pharmacopoeia, then write, "Complies the test"; if not then write "Does not comply the test".

10. Result

The given sample of aspirin complies with the tests _____ and does not comply with the tests _____for identification as per IP 2022.

11. Conclusion

Identification tests for aspirin were performed as per the procedure given in IP 2022.

12. Reference

Indian Pharmacopoeia 2022, Volume II.

13. Practical Related Questions

a. Draw the structure of aspirin.
b. Write the chemical name of aspirin.

c. Give storage condition of aspirin.
d. Mention two brand names of aspirin.
e. Write any one chemical identification test of aspirin.

(*Space for Answers*)

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14. Assessment Scheme

Particular	Understanding the basic concept (Intellectual skill)	Activities (Motor skill)	Cleanliness & Handling (Affective domain)	Viva-voce / Answers Written	Total	Signature of teacher
Marks Obtained						
Max Marks	**02**	**05**	**01**	**02**	**10**	

Experiment No. 20
Identification test of Caffeine

1. Aim
To perform and report identification tests on the given sample of Caffeine as per IP-2022.

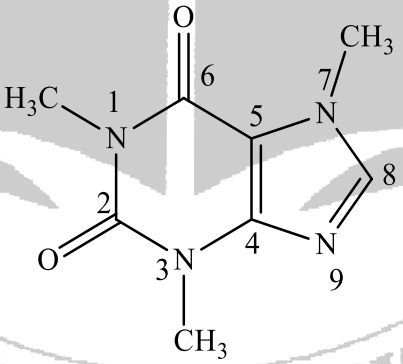
2. Practical Significance
The purpose of identification tests is to ensure accurate labeling of materials. Identification typically involves a combination of straightforward chemical tests and measurement of relevant physical constants. Chemical tests used for identification aim to establish the presence of specific functional groups, thereby confirming the molecular structure as accurately as possible. Inorganic substances generally rely on tests commonly used in qualitative analysis. Organic substances are identified through characteristic reactions of the functional groups present in their molecules.

3. Practical Outcomes
After completion of this practical, the students will be able to:

PrO	Practical Outcomes	Mapped CO	BTL
1	Describe the monograph of caffeine.	CO5	2
2	Perform identification tests on the given sample for caffeine as per IP.	CO5	5
3	Write a report on the identification test.	CO5	3
4	Follow cleanliness, safety and ethical practices.	CO5	5

4. Relevant Theoretical Background
Monograph of Caffeine as per IP 2022

Caffeine is 3, 7-dihydro-1, 3, 7-trimethyl-1*H*-purine-2, 6-dione or its monohydrate.



Molecular formula	$C_6H_{10}N_4O_2$
Molecular weight	194.2g (anhydrous)
Molecular formula	$C_6H_{10}N_4O_2, H_2O$
Molecular weight	212.2g (monohydrate)
Standard	Caffeine contains not less than 98.5 percent and not more than 101.5 percent of $C_6H_{10}N_4O_2$ calculated on the dried basis.
Organoleptic description	**Nature**- Silky crystals, glistering needles, or crystalline powder **Colour**- White **Odour-** Odourless **Taste**- Bitter

Solubility	Sparingly soluble-water and alcohol Freely soluble- Chloroform Soluble- Ether
Pharmaceutical category	Central nervous system stimulant
Storage	Store protected from light and moisture
Labelling	The label states whether it is anhydrous or monohydrate.
Dose	300 to 600mg

5. Requirements

a. Glasswares: Porcelain dish, Thiele's tube, Capillary, Test tubes, Glassrod

b. Chemicals: Hydrochloric acid, Potassium chlorate, Dilute ammonia solution, Tannic acid, Iodine, Dilute sodium hydroxide

c. Reagents

a) 0.05 M iodine: Dissolve about 14 g of iodine in a solution of 36 g of potassium iodide in 100 ml of water. Add three drops of hydrochloric acid and dilute with water to 1000 mL.

6. Requirements used

_____

_____

7. Precautions

a. Use clean and dry glass apparatus.

b. Use acid-resistant materials such as glass or plastic when handling tannic acid. Avoid using reactive materials such as metals, which may react with the acid.

8. Procedure

a. **Organoleptic description**

Observe the given drug critically for the following description.

The drug is silky white crystals, white glistering needles, or white crystalline powder, odourless as per I.P. 2022

b. **Solubility test**

Perform solubility tests in the different solvents. The drug is sparingly soluble in water and in alcohol, freely soluble in chloroform, and slightly soluble in ether.

c. **Identification test**

Test A. To 10 mg in a porcelain dish, add 1mL of hydrochloric acid and 0.1 g of potassium chlorate and evaporate to dryness on a water bath. Exposure of the residue to the vapours of dilute ammonia solution; a purple colour is produced which disappears with the addition of a solution of a fixed alkali.

Test B. To the saturated solution, add a few drops of tannic acid solution; a white precipitate which is soluble in excess of the reagent.

Test C. To 5 mL of saturated solution add 1.5 mL of 0.05m iodine, the solution remains clear. Add a few drops of dilute hydrochloric acid; a brown precipitate is formed which dissolves on neutralization with sodium hydroxide solution.

Test D. Determination of melting point: The drug melts at about 235^0C-239^0C.

9. Observations

Identification test report of the sample of Caffeine

Sr. No.	Test	Observation	Inference*
1. a)	**Organoleptic description** Nature		
b)	Colour		
c)	Odour		
2. a)	**Solubility** Water		
b)	Alcohol		
c)	Chloroform		
d)	Ether		
3. a)	**Identification Test** Test A		
b)	Test B		
c)	Test C		
d)	Test D		

*If observation is as per given Indian Pharmacopoeia, then write, "Complies the test"; if not then write "Does not comply the test".

10. Result

The given sample of caffeine complies with the tests _____ and does not comply with the tests _____for identification as per IP 2022.

11. Conclusion

Identification tests were performed as per the procedure given in IP 2022.

12. Reference

Indian Pharmacopoeia 2022.

13. Practical Related Questions

a. Define Analeptics.
b. Enlist two marketed preparations of caffeine.
c. Draw the structure of caffeine and give its chemical name.
d. Name the heterocyclic ring present in the caffeine. Draw its structure.
e. Write any one chemical identification test of caffeine.

(*Space for Answers*)

14. Assessment Scheme

Particular	Understanding the basic concept (Intellectual skill)	Activities (Motor skill)	Cleanliness & Handling (Affective domain)	Viva-voce / Answers Written	Total	Signature of teacher
Marks Obtained						
Max Marks	**02**	**05**	**01**	**02**	**10**	

Experiment No. 21
Identification test of Paracetamol

1. Aim
To perform and report identification tests on the given sample of Paracetamol as per IP-2022.

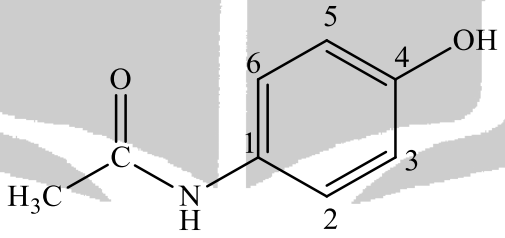
2. Practical Significance
The purpose of identification tests is to ensure accurate labeling of materials. Identification typically involves a combination of straightforward chemical tests and measurement of relevant physical constants. Chemical tests used for identification aim to establish the presence of specific functional groups, thereby confirming the molecular structure as accurately as possible. Inorganic substances generally rely on tests commonly used in qualitative analysis. Organic substances are identified through characteristic reactions of the functional groups present in their molecules.

3. Practical Outcomes
After completion of this practical, the students will be able to:

PrO	Practical Outcomes	Mapped CO	BTL
1	Describe the monograph of paracetamol.	CO5	2
2	Perform identification tests on the given sample for paracetamol as per IP.	CO5	5
3	Write a report on the identification test.	CO5	3
4	Follow cleanliness, safety and ethical practices.	CO5	5

4. Relevant Theoretical Background
Monograph of paracetamol as per IP 2022

Paracetamol is, chemically named as 4-hydroxyacetanilide.



Molecular formula	$C_5H_9NO_2$
Molecular weight	151.2g
Standard	Paracetamol contains not less than 99.0 percent and not more than 101.0 percent of $C_9H_8O_4$ calculated on the dried basis.
Organoleptic description	**Nature**- Crystals, crystalline powder **Colour**- White **Odour-** Odourless **Taste**- Slightly bitter
Solubility	Freely soluble- Alcohol (95%), acetone, and solution of alkali hydroxide Sparingly soluble- Water Very slightly soluble- Ether

Pharmaceutical category	Analgesic ; antipyretic
Storage	Store protected from light and moisture
Dose	500 to 1 g every 4 to 6 hours, up to 4 g daily, in divided doses

5. Requirements

a. Glassware: Thiele's tube, Capillary, Test tubes, Glassrod

b. Chemicals: Sodium hydroxide, Ether, Ferric chloride, Hydrochloric acid, Potassium dichromate, Acetone, Alcohol.

6. Requirements used

_____

_____

7. Precautions

a. Use clean and dry glass apparatus.

b. When sodium hydroxide comes into contact with the skin, it can trigger severe irritation and burns, potentially causing harm to both the skin and eyes. Additionally, prolonged exposure might lead to the accumulation of fluid in the lungs, a condition is known as pulmonary edema, which requires urgent medical attention.

8. Procedure

a. **Organoleptic description**

Observe the given drug critically for the following description.

The drug is white crystals, white glistering needles, or white crystalline powder, odourless as per I.P. 2022

b. **Solubility test**

Perform solubility tests in the different solvents. The drug is sparingly soluble in water, freely soluble in alcohol (95%), acetone, and solution of alkali hydroxide, and very slightly soluble in ether.

c. **Identification test**

Test A. Dissolve 0.1 g of sample in 10 mL of water and add 0.1 mL of ferric chloride solution; a violet colour develops.

Test B. Boil 0.1 g in 1 mL of hydrochloric acid for 3 minutes, add 10 mL of water, and cool; no precipitate is produced. Add 0.05 mL of 0.0167 m potassium dichromate; a violet colour develops which does not turn red. (Distinction from phenacetin)

Test C. Determination of the melting point: The drug melts at about 168^0C-172^0C.

9. Observations

Identification test report of the sample of Paracetamol

Sr. No.	Test	Observation	Inference*
1. a)	**Organoleptic description** Nature		
b)	Colour		

Sr. No.	Test	Observation	Inference*
c)	Odour		
2. a)	**Solubility** Water		
b)	Alcohol		
c)	Chloroform		
d)	Ether		
3. a)	**Identification Test** Test A		
b)	Test B		
c)	Test C		

*If observation is as per given Indian Pharmacopoeia, then write, "Complies the test"; if not then write "Does not comply the test".

10. Result

The given sample of Paracetamol complies with the tests _____ and does not comply with the tests _____for identification as per IP 2022.

11. Conclusion

Identification tests for Paracetamol were performed as per the procedure given in IP 2022.

12. Reference

Indian Pharmacopoeia 2022.

13. Practical Related Questions

a. Draw the structure and write IUPAC name of paracetamol.
b. Give storage condition of paracetamol.
c. Mention two popular brand names of paracetamol.
d. Write any one chemical identification test of paracetamol.
e. What is the dose of paracetamol?
f. List uses of paracetamol.

(*Space for Answers*)

14. Assessment Scheme

Particular	Understanding the basic concept (Intellectual skill)	Activities (Motor skill)	Cleanliness & Handling (Affective domain)	Viva-voce / Answers Written	Total	Signature of teacher
Marks Obtained						
Max Marks	**02**	**05**	**01**	**02**	**10**	

Experiment No. 22
Identification test of Sulphanilamide

1. Aim
To perform and report identification tests on the given sample of Sulphanilamide as per IP.

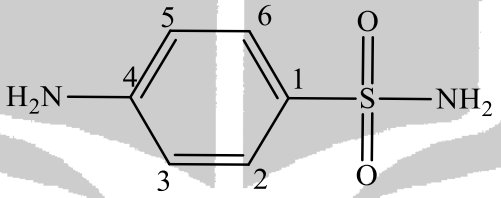
2. Practical Significance
The purpose of identification tests is to ensure accurate labeling of materials. Identification typically involves a combination of straightforward chemical tests and measurement of relevant physical constants. Chemical tests used for identification aim to establish the presence of specific functional groups, thereby confirming the molecular structure as accurately as possible. Inorganic substances generally rely on tests commonly used in qualitative analysis. Organic substances are identified through characteristic reactions of the functional groups present in their molecules.

3. Practical Outcomes
After completion of this practical, the students will be able to:

PrO	Practical Outcomes	Mapped CO	BTL
1	Describe the monograph of sulphanilamide.	CO5	2
2	Perform identification tests on the given sample for sulphanilamide as per IP.	CO5	5
3	Write a report on the identification test.	CO5	3
4	Follow cleanliness, safety and ethical practices.	CO5	5

4. Relevant Theoretical Background
Monograph of Sulphanilamide as per I.P.

Sulphanilamide is, chemically named 4-aminobenzenesulphonamide.



Molecular formula	$C_6H_8N_2O_2S$
Molecular weight	172.2g
Standard	Sulphanilamide contains not less than 99.0 percent and not more than 105.0 percent of $C_6H_8N_2O_2S$ calculated on the dried basis.
Organoleptic description	**Nature**- Crystalline solid powder **Colour**- White **Odour-** Odourless **Taste**- Slightly bitter
Solubility	Soluble- Hot water, alcohol and acetone Insoluble- Chloroform, ether, and petroleum ether
Pharmaceutical category	Antibacterial

Storage	It may be unstable if exposed to long periods of air and light, hence stored in a cool and dry place.

5. Requirements

a. Glassware: Thiele's tube, Capillary, Test tubes, Glass rod

b. Chemicals: Sodium hydroxide, Ethanol, Ether, Acetone, Sodium nitrite, Dil. hydrochloric acid, β-naphthol.

6. Requirements used

_____

_____

7. Precautions

a. Use clean and dry glass apparatus.

b. Acetone, chloroform and ether, the solvents for the solubility test are very volatile and flammable. No flames will be allowed in the lab once ether is being used.

8. Procedure

a. **Organoleptic description**

Observe the given drug critically for the following description.

The drug is **a** white crystalline powder, odourless as per I.P.

b. **Solubility test**

Perform solubility tests in the different solvents. The drug is soluble in hot water, alcohol, and acetone water and insoluble in chloroform, ether, and petroleum ether

c. **Identification test**

Test A. Dissolve 0.1 g of sample in 2 ml of water and add 2 mL of 10% NaOH solution. Heat this solution, ammonia evolves.

Test B. Dissolve 0.1 g of sample in 2 mL dil. HCl and cool the mixture to 0^0 C. To this solution add precooled 2 ml of 2% $NaNO_2$ solution. Add 1 mL of this solution to a solution of β-naphthol in NaOH; orange red or red colour dye is obtained.

Test C. Determination of the melting point: The drug melts at about 165^0C.

9. Observations

Identification test report of the sample of Sulphanilamide

Sr. No.	Test	Observation	Inference*
1. a)	**Organoleptic description** Nature		
b)	Colour		
c)	Odour		
2. a)	**Solubility** Water		
b)	Alcohol		

Sr. No.	Test	Observation	Inference*
c)	Chloroform		
d)	Ether		
3. a)	**Identification Test** Test A		
b)	Test B		
c)	Test C		

*If observation is as per given Indian Pharmacopoeia, then write, "Complies the test"; if not then write "Does not comply the test".

10. Result

The given sample of Sulphanilamide complies with the tests _____ and does not comply with the tests _____for identification as per IP.

11. Conclusion

Identification tests for Sulphanilamide were performed as per the procedure given in IP.

12. Reference

Indian Pharmacopoeia 2022.

13. Practical Related Questions

a. Draw the structure of sulphanilamide.
b. Write the chemical name of sulphanilamide.
c. Give storage condition of sulphanilamide.
d. Mention two popular brand names of sulphanilamide.
e. Write any one chemical identification test of sulphanilamide.

(*Space for Answers*)

14. Assessment Scheme

Particular	Understanding the basic concept (Intellectual skill)	Activities (Motor skill)	Cleanliness & Handling (Affective domain)	Viva-voce / Answers Written	Total	Signature of teacher
Marks Obtained						
Max Marks	**02**	**05**	**01**	**02**	**10**	

SYSTEMATIC QUALITATIVE ANALYSIS

The analysis and identification of unknown organic compounds founds a very important aspect of experimental pharmaceutical chemistry. Often, a common first step in the identification of organic compounds is to detect elements present in given sample. In this analysis students will carry out several qualitative tests that will allow them to identify functional groups in organic compounds. There is no certain set procedure that can be generally applied to organic qualitative analysis. Various books/literature have different methods, but a systematic method based on the scheme given below will give good results.

The systematic qualitative analysis of organic compounds includes the following different steps.

1. **Preliminary Tests**
   In these tests, students can note all the physical characteristics of the compound that includes solid, liquid, colour or odour. Also, performing an ignition test by heating a small amount of metal spatula helps in determining whether the compound is aromatic or aliphatic as aliphatic compounds produce luminous flame and aromatic compounds produce a sooty flame. Solubility test is performed using various reagents such as dil. HCl, dil. NaOH, sodium bicarbonate and hot or cold water. Very useful information can be obtained by observing the organic compound's solubility.

2. **Determination of Physical Constant**:
   A pure organic compound usually has a sharp and definite melting point which is characteristic of that compound. The accurate determination of melting or boiling point of organic compounds plays a very important role in its identification.

3. **Detection of elements (Elemental Analysis)**
   Organic compounds have been classified into various types, depending upon elements present in them. Hence the detection of elements present in the compound is important for identifying the class of organic compounds. Elemental analysis is finding out all elements present in the organic compound by some color and precipitation reaction.

4. **Detection of Functional Group**
   A functional group is the group of atoms present in the compounds that are responsible for the chemical reaction of those compounds. Group analysis is to find out different functional groups present in the organic compound.

5. **Identification of the compound/drug by search of literature with similar physical and chemical properties.**
   It is a reference table in which compounds are classified according to elements, groups and physical constants. The final identification of compounds is done with reference to this information.

6. **Confirmative Test (Specific colour reaction or preparing suitable derivative.)**

   The identity of organic compounds is finally confirmed by the specific colour reactions or preparation of simple derivatives and determining its M. P. / B. P.

1. Preliminary Tests and physical examination:

S No	Test	Observation	Inference
1.	**Physical State**	Solid	Carbohydrate, acid, phenol, amine, higher hydrocarbon may be present
		Liquid	Alcohol, ketone, aldehyde, ester, phenol, amines may be present.
2.	**Colour**	Colorless Solid	Simple acid, alcohol, ester, aromatic hydrocarbon, ketone.
		Colorless Liquid	Alcohols, aldehydes, ketones, esters, ethers, aromatic hydrocarbons, etc
		Yellow - Solid	m- Dinitrobenzene, p- Nitro toluene, nitro phenol, nitro aniline.
		Yellow - liquid	Nitrobenzene.
		Brown	P – Toluidine, resorcinol.
		Blackish	α – Naphthol
		Pink	ß – Naphthol
3.	**Odour**	Sweet, pleasant	Ester, alcohol, and halogen derivatives
		Fishy	Amines
		Phenolic	Phenols, Naphthols
		Fruity	Esters
		Deep Sweet	Chloroform
		Odour of bitter	Nitrobenzene, Nitrotoluene
		Almond	Benzaldehyde
		Cucumber like odour	Chloral or Chloral hydrates
		Pungent & irritating odour	Acid halides, Acetic acid, Formaldehyde.
4.	**Solubility**		
	Take about 0.1 g of solid or 0.2 mL of liquid sample in a test tube and treat it with about 3 mL of solvent. Shake well. Warm if necessary and cool to room temperature and observe.		
	A) Soluble in water	Solution is tested with litmus	
	Litmus Paper test **If acidic,** add substance to 10% Sodium Bicarbonate solution.	Blue litmus paper turns red	Acid or phenol is present
		Evolution of carbon dioxide with effervescence	Carboxylic acid, acid salts may present.
		No Evolution of carbon dioxide	Phenols may present.
	If nonacidic – perform a red litmus paper test.	Red litmus paper turns blue	Base is present
		No change Blue / red litmus paper	Neutral compound is present

S No	Test	Observation	Inference
			(Alcohols, carbohydrates, amides, ketones, aldehydes)
	B) Insoluble in water		
	i. Sub + 10% $NaHCO_3$.	Soluble with strong effervesces.	Carboxylic acid, acid salts may present.
		Reprecipitated by adding Conc. HCl (drop by drop)	Carboxylic acid confirmed.
	ii. Sub + 10% NaOH	Soluble	Phenols may present.
		Reprecipitated by adding Conc. HCl (drop by drop)	Phenols confirmed.
	iii. Sub + Dil HCl	Soluble	Base may Present (amines)
		Reprecipitated by adding 20% NaOH (drop by drop)	Base confirmed.
	iv. If the substance is insoluble in $NaHCO_3$, NaOH, HCl.		Neutral compound is present.
	Sub + cold conc. H_2SO_4	Soluble	Ethers, esters, aromatic hydrocarbons, etc may present
		Insoluble	Hydrocarbons, Halogen Derivatives of hydrocarbons.
5.	**Action of Reagents**		
i. **Action of cold NaOH** Sub + 2 mL Water + 2mL 10% NaOH		i. Evolution of ammonia ii. Change in colour a. Yellow to orange red b. Colourless to deep yellow c. Gives blue – black colour	Ammonium salts o-nitrophenols m- and p- nitrophenols polyhydroxy phenols, amino phenols, benzoquinone.
ii. **Action of Hot NaOH** Warm the above mixture strongly		i. Evolution of ammonia ii. Evolution of Chloroform iii. Brown Colour iv. Darkness v. Odour of alcohols vi. Yellowish green colour vii. Yellow colour	Amides e.g. Urea, thiourea Choral, chloral hydrates Pyrogallol Carbohydrates, polynitro Esters of aliphatic alcohols Resorcinol Glucose
iii. **Action of Hot Conc. H_2SO4** Sub + 1 mL conc. H_2SO_4, warm		i. Blackening with effervescence of CO, CO2, SO2 ii. Blackening without effervescence iii. Effervescence but no blackening iv. No effervescence no blackening but pungent odour	Carbohydrates as cane sugar, higher hydroxy acids Polyphenols, Phenolic acids Formic, citric, oxalic acid Acetic, benzoic, succinic acids, esters, phenols

S No	Test	Observation	Inference
		v. Yellow to brown but no charring.	Glucose
iv.	**Action of Na_2CO_3** Sub + 5 mL 10%. Na_2CO_3 solution	Effervescence	Acids may Present
v.	**Test for unsaturation** **a. Baeyer's Test** Sub + 5 mL 10%. Na_2CO_3 solution + drop by drop $KMnO_4$ solution	Decolorization of $KMnO_4$	Unsaturated compounds.
		No decolorization of $KMnO_4$	Saturated compound
	b. Bromine Water Test Sub + 2 mL water + 2 mL bromine water shake well	Decolorization without ppt	Unsaturated compounds
		Decolorization with ppt	Phenols, aromatic amines
		No Decolorization	Saturated compounds.
vi.	**Action of $FeCl_3$ solution** Sub + 2 mL water + 2 mL $FeCl_3$ solution, shake well	Green coloration changing through blue and violet, red by addition of Na_2CO_3	Catechol
		Blue or violet coloration	Phenols, Phenolic acids
		Red Coloration	Pyrogallol, Guaiacol
vii.	**Heating on Copper Gauze** Small copper foil and heat it in the flame. Place 0.2 g sample on it and heat in the flame.	Sooty flame	Aromatic compound or aliphatic compound containing small proportion of hydrogen e.g., $CHCl_3$, CCl_4
		Non sooty flame	Aliphatic compounds
		Charring and smell of burnt sugar	Carbohydrate, sulphanilic acid.
viii.	**Heating with Soda Lime** Sub + 2g finely powdered soda lime + 1 g coarse soda lime and heat	Evolution of ammonia	Amides, thiourea,
		Smell of caramel	Carbohydrates
		Benzene smell	Aromatic carboxylic acid, Benzoic acid.

Conclusion: On the basis of the tests performed above the given organic compound is

1. Aromatic / Aliphatic
2. Saturated / Unsaturated
3. Acidic / Phenolic / Basic / Neutral in nature.

1. DETERMINATION OF PHYSICAL CONSTANT

Conclusion: The melting / boiling point of a given organic compound was found to be _____.

2. DETECTION OF ELEMENTS

Lassaigne's Test / Sodium Fusion Test: Principle

Nitrogen, Sulphur, and halogens present in organic compounds are detected by Lassaigne's test. This test is based on fusion of sodium metal ions with the elements present in organic compounds converted into ionic salts. If nitrogen is present in organic compounds, then it gets converted to cyanide ions.

Similarly, sulphur and halogens present in organic compounds are converted to sulphide and halide ions respectively. These ionic salts are then tested in the usual manner.

Reaction:

$$Na + C + N \longrightarrow NaCN$$

Sodium cyanide

$$N + 2Na \longrightarrow Na_2S$$

Sodium sulphide

$$X(Cl, Br \text{ or } I) + Na \longrightarrow NaX$$

Sodium halide

Sodium fusion extract is treated with ferrous sulphate in presence of base, it forms sodium ferrocyanide. By the action of sulphuric acid, the sodium ferrocyanide reacts with ferric sulphate producing ferric ferro cyanide that is prussian blue in colour confirming presence of nitrogen.

$$FeSO_4 + 6NaCN \xrightarrow{NaOH} Na_4[Fe(CN)_6] + Na_2SO_4$$

Sodium ferrocyanide

$$3Na_4[Fe(CN)_6] + 2Fe_2(SO_4)_3 \longrightarrow Fe_4[Fe(CN)_6]_3 + 6Na_2SO_4$$

Ferric ferro cyanide

Sodium sulphide, from sodium fusion test react with sodium nitroprusside to produce violet coloration indicating presence of sulphur.

$$Na_2S + Na_2[Fe(CN)_5NO] \longrightarrow Na_4[Fe(CN)_5NOS]$$

Sodium nitroprusside — Violet colour

In another reaction sodium sulphide reacts with lead acetate resulting in the formation of black PPT of lead sulphide, indicating presence of sulphur.

$$Na_2S + (CH_3COO)_2Pb \longrightarrow PbS\downarrow + 2CH_3COONa$$

Lead acetate — Lead sulphide

Preparation of sodium fusion extract:

In a dry fusion tube, insert a piece of sodium metal and heat gently till sodium melts. Add the same amount of sample into the fusion tube. Slowly melt the content and allow the sample to react with sodium slowly. Heat the sodium fusion tube to red hot and drop this tube in water (approximately 10 mL/fusion tube) placed in the porcelain dish, simultaneously covered by wire gauze held in the left hand. With the help of a glass rod, break further pieces of sodium fusion tubes to extract the organic salts from the fused mass. Heat the content to boiling and concentrate the mixture to half the volume. Filter the mixture. Filtrate is known as sodium extract. Use this filtrate for the following test.

S.No	Test	Observation	Inference
1.	**Test for Nitrogen** 2 mL of extract + a few drops of freshly prepared solution of $FeSO_4$; green precipitate is obtained. If not obtained, add NaOH solution and boil the mixture a few minutes, cool and acidify it with by adding dil.HCl or dil. H_2SO_4.	Prussian Blue Colour appears	Nitrogen present
2.	**Test for Sulphur** 2 mL of extract + 2-3 drops of freshly prepared sodium nitroprusside solution.	Intense purple colour	Sulphur present
	2 mL of extract +acidify with dilute Acetic Acid + 1 mL of Lead Acetate Solution.	Black precipitation	Sulphur present
3.	**Test for nitrogen and sulphur together** 2 mL of extract + dilute HCl + 2-3 drops of $FeCl_3$ solution.	Red or Blood -Red colour	Nitrogen and Sulphur present
4.	**Test for Halogens** (Precaution: Rinse test tube with distilled water) 1 mL of extract +1 mL of dilute HNO_3 (boil well if N and S are present) + 1 mL of 5% $AgNO_3$ solution.	White precipitate which dissolves on addition of NH_4OH	Chlorine present
		Yellowish white precipitate which partially dissolves on addition of NH_4OH	Bromine present
		Yellow precipitate insoluble in NH_4OH	Iodine present
	Separation of Cl, Br and I If halogen is present carry out the following test: 1mL of extract + 1mL of dilute H_2SO_4 / HNO_3 boil & cool. Add 1 mL of $CHCl_3$ or CCl_4 and chlorine water in excess, shake well and observe the colour of the chloroform layer.	Colorless	Chlorine present
		Yellow orange or reddish brown	Bromine present
		Violet	Iodine present

Conclusion: The given organic compound contains _____ elements.

3. DETECTION OF FUNCTIONAL GROUP

Based on above conclusions, organic compounds are grouped into four groups which may be further divided in subgroups

Group	Elements present in organic compound	Group	Elements present in organic compound
I	**C, H, (O) Elements**	**III**	**C, H, (O), N and S Elements**
	A. Acidic		A. Acidic
	B. Phenolic		B. Phenolic
	C. Basic		
II	**C, H, (O) and N Elements**	**IV**	**C, H, (O) and Halogens**
	A. Acidic		Neutral
	B. Phenolic		
	C. Basic		
	D. Neutral		

Group I: Compounds containing C, H, (O) elements			
Group I: A - Tests for acids: Compounds may contain carboxylic acid (-COOH) functional groups			
S.No	**Test**	**Observation**	**Inference**
1.	Sub + 5 mL of saturated solution of sodium bicarbonate	Soluble with strong effervesces of CO_2.	Carboxylic acid, acid salts may present.
		Reprecipitated by adding Conc. HCl (drop by drop)	Carboxylic acid confirmed.
2.	**Neutral test Solution** 1 g of sample + 1 mL water + phenolphthalein solution + Ammonia solution till just alkaline. Now boil to remove excess of ammonia (drop of solution no longer turns red litmus blue) and add $FeCl_3$ drop by drop.	Buff colored ppt	Benzoic or phthalic acid
		Violet-colored ppt.	Salicylic acid
		Faint reddish coloration or ppt.	Acetyl salicylic acid.
		Yellow-colored ppt	Cinnamic acid
3.	Neutral solution from test 2 + $CaCl_2$ solution	White ppt in cold insoluble in acetic acid	Oxalic acid
		White ppt in cold soluble in acetic acid	Tartaric acid
		White ppt only on boiling insoluble in acetic acid	Citric acid or Malic acid
		White ppt only on boiling soluble in acetic acid	Succinic acid
Examples of acidic compounds with literature M.P. / B. P.			

Name	**Physical Constant**	**Confirmatory test/ Derivatives**
Benzoic acid	M. P. 121 – 122°C	a. Neutral test solution – buff colored ppt b. Anilide derivative (m.p. 162 °C) c. p-toluidine derivative (m.p. 158 °C) d. Amide derivative (m.p. 129 °C)
Salicylic acid	M. P. 158°C	a. Neutral test solution – violet colored ppt b. Anilide derivative (m.p. 135 °C) c. p-toluidine derivative (m.p. 156 °C) d. Amide derivative (m.p. 139 °C)
Acetyl salicylic acid	M. P. 135 °C	a. Neutral test solution – Faint reddish color or ppt b. 2 g acid + 2 mL dil. NaOH, boil for 5 min. Coll and acidify with dil. HCl, white ppt of salicylic acid (m. p. 156°C c. Anilide derivative (m.p. 136 °C) d. Amide derivative (m.p. 138 °C)

Group I: B - Tests for Phenols: Compounds may contain phenolic (-OH) functional group			
S.No	**Test**	**Observation**	**Inference**
1.	Sub + 10% NaOH	Soluble	Phenolic (-OH) group may present.
		Reprecipitated by adding Conc. HCl (drop by drop)	Phenols confirmed.
2.	**Action of $FeCl_3$ solution** Sub + 2 mL water or alcohol + 2 mL $FeCl_3$ solution, shake well	Violet colour produced	Phenol is present
		Violet-blue or blue coloration	Resorcinol, Cresol may present
		Greenish white opalescent.	β – naphthol may present
		Blue- violet	α – naphthol may present
3.	**Phthalein Test** Sub + 0.2 g phthalic anhydride + few drops of conc. H_2SO_4. Warm, cool Pour it into a beaker containing dil. NaOH solution.	Pink - Red colour	Phenol or cresol
		Yellow – green fluorescene	Resorcinol may present
		Faint green colour	α – naphthol may present
		Green or bluish green	β – naphthol may present
4.	**Libermann's Reaction** Sub + few crystals of solid $NaNO_2$ + few drops of Conc. H_2SO_4	Green or blue colour produced and change to red on dilution.	Phenolic (-OH) group is present.

Examples of phenolic compounds with literature M.P. / B. P.		
Name	**Physical Constant**	**Confirmatory test/ Derivatives**
Resorcinol	M. P. 110°C	a. Phthalein test – Yellow-green fluorescence produced. b. Bromo derivative, m. p. 112°C c. Benzoate derivative, m. p. 117°C
α – naphthol	M. P. 95°C	a. Aqueous solution gives white ppt with FeCl3 b. Sub + 2 mL of aqueous NaOH + drop of CCl4 + pinch of copper powder, warm, blue colour produced. (Distinction for β – naphthol) c. Bromo derivative, m. p. 105°C d. Acetate derivative, m. p. 49°C e. Benzoate derivative, m. p. 56°C
β – naphthol	M. P. 123°C	a. Sub + 2 mL of aqueous NaOH + drop of CCl4 + pinch of copper powder, warm; not produced any colour. b. Bromo derivative, m. p. 84°C c. Acetate derivative, m. p. 72°C d. Benzoate derivative, m. p. 107°C
Catechol	M. P. 105°C	a. Sub + 2 mL of water + 2 mL lead acetate solution; white ppt produced. b. Bromo derivative, m. p. 192°C c. Acetate derivative, m. p. 65°C d. Benzoate derivative, m. p. 84°C

Group I: C – Test for Neutral compounds

These compounds may contain one or more functional group such as aldehyde, ketone, alcohol, ester, ethers, hydrocarbons, carbohydrates.

S.No	Test	Observation	Inference
1.	**Test for Aldehyde** A. Sub + 5 mL dil. HCl _ 2 mL of 2,4-dinitrophenyl hydrazine in dil. HCl. Cool and allow to stand for few min.	Yellow, orange or red-coloured crystalline precipitate	Aldehyde (-CHO) or Ketone (-C=O) functional group is present.
	B. **Schiff's Test** Sub + 2 mL of Schiff's reagent, shake for 2 min.	Deep violet-red or red colour.	Aldehyde (-CHO) group is present.
	C. **Tollen's Test (Silver – Mirror Test)** Sub + 2 mL of Tollen's reagent. Heat it in boiling water bath for 5 min	Silver mirror deposits on the walls of test tube.	Aldehyde (-CHO) group is present.
	D. **Fehling's Test** 1 mL of Fehling A + 1 mL of Fehling B solution + Sub and boil for 5 min.	Reddish brown precipitate of Cuprous oxide	Aldehyde (-CHO) group is present.
2.	**Test for Ketone** Sub + 2 mL freshly prepared sodium nitroprusside solution + few drops of NaOH solution	Wine – red or orange red	Ketonic (-C=O) group is present.
Test for Alcohols			
3.	**Test for Alcohols** A. **Ceric Ammonium Nitrate test** 1 mL substance + few drops of ceric ammonium nitrate reagent	Red colour	Alcoholic –OH group is present.
	B. Sub + 2-3 drops of acetyl chloride, HCl gas evolved. Glass rod is dipped in NH_4OH brought in contact with HCl gas	White fumes produced	Alcoholic –OH group is present.
	C. In china dish 0.5 mL Sub + 1 mL benzene + small piece of sodium metal	Effervescences of hydrogen gas	Alcohol Present
	D. **Lucas Test** To 0.2 mL or 0.2 g of the compound in a test tube add 2 mL of the Lucas reagent and shake well	**a**. Cloudy layer separated immediately **b**. Cloudy layer separated 5-10 minutes **c**. A clear homogeneous solution	Tertiary alcohol Secondary alcohol Primary alcohol
4.	**Test for Esters** **A.** Sub + 10 mL alcohol + few drops of NaOH + drop of phenolphthalein + Heat the reaction mixture for 5 min.	Disappearance of Pink colour	Ester group is present
	B. Hydroxamic acid test (Feigl) Sub + 0.2 g solid hydroxylamine hydrochloride + 5 mL NaOH	Violet or deep red colour	Ester group is present

S.No	Test	Observation	Inference
	solution. Boil, cool, acidify with dilute HCl, add few drops of $FeCl_3$		
5.	**Tests for Ethers** **A.** 2 to 3 drops of sub + 5 mL of benzene + 5 mL of very dilute solution of iodine in benzene, shake well.	Brown tint colour	Ethers present.
	B. 2 mL of sub in boiling tube, cover the mouth of tube with a filter paper moistened with a mixture of cupric acetate and benzidine hydrochloride. Heat the tube for 3-5 minutes.	Deep – blue colour appears on filter paper	Ether group is present.
	C. Hydroxamic acid test (Feigl) Sub + 0.2 g solid hydroxylamine hydrochloride + 5 mL NaOH solution. Boil, cool, acidify with dilute HCl, add few drops of $FeCl_3$	Violet or deep red colour	Ether group is present
6.	**Tests for Carbohydrates**		
	i. Molisch's Test Sub + water + 2 drops of Molisch's Reagent. Shake well and add 2 mL conc. H_2SO_4 along the side of test tube.	Formation of reddish violet ring at the junction of two liquids. Which on shaking produces deep violet solution	Carbohydrate may present.
	ii. Water Solubility Test for Carbohydrates Sub + water	Water soluble	Glucose, fructose, galactose, lactose and maltose
		Water insoluble	Sucrose and insulin
	iii. Barfoed's Test 1 mL carbohydrate solution + 3 mL Barfoed's reagent heat in Boling water bath	Red precipitate within two minutes	Presence of monosaccharides ie glucose, fructose, galactose.
		Red precipitate after ten minutes	Presence of disaccharides i.e. Lactose or maltose
	iv. Rapid Furfural Test: Sub + water + few drops of Molisch Reagent. Shake well and add 3 mL conc. HCl boil the solution.	Violet colour within 30 sec	Fructose or Sucrose
		Violet colour after 1 min or no violet colour.	Glucose
	v. Seliwanoff's test Sugar + water + add 10 drops of seliwanoff's reagent and heat the mixture to boiling. Note: If heating is prolonged aldose will also develop the colour.	Cherry red colour develops within 2 minutes	Indicates the presence of ketose such as fructose.

S.No	Test	Observation	Inference
7.	**Test for Hydrocarbons** **i. Iodine Test** 2 to 3 drops of sub + 5 mL of benzene followed by very dilute solution of iodine in benzene	Solution remains violet in colour	Hydrocarbon present
	ii. Friedel Craft condensation test: Take 0.5 g of anhydrous AlCl3 in dry test tube, heat. When AlCl3 sublimes to deposit on upped end of tube, add 2-3 drops of mixture of equal amounts of substance and CHCl3.	Orange, Red, Blue, or green colour due to formation of triphenylmethane dyes.	Aromatic hydrocarbons present.

Examples of neutral compounds with literature M.P. / B. P.		
Name	**Physical Constant**	**Confirmatory test/ Derivatives**
Benzaldehyde	B. P. 179ºC	Sub + 2 mL alkaline $KMnO_4$, heat, filter and acidify the filtrate with dil. HCl. A white precipitate of benzoic acid is obtained. 2,4, Dinitrophenylhydrazone derivatives m. p. 237ºC Semicarbazone derivative, m.p. 224ºC Oxime derivative, m.p. 35ºC
Salicylaldehyde	B. P. 197ºC	Sub + 2 mL alkaline $KMnO_4$, heat, filter and acidify the filtrate with dil. HCl. A white precipitate of salicylic acid is obtained. 2,4, Dinitrophenylhydrazone derivatives m. p. 252ºC Semicarbazone derivative, m.p. 231ºC Oxime derivative, m.p. 63ºC
Acetone	B. P. 56ºC	Iodoform test: Sub + few drops of iodine solution + NaOH solution, warm, brown colour of iodine disappears and a yellow precipitate of iodoform is formed (hospital like smell) 2,4, Dinitrophenylhydrazone derivatives m. p. 128ºC Semicarbazone derivative, m.p. 190ºC Oxime derivative, m.p. 59ºC
Dextrose	M. P. 146ºC	1 g Lead acetate + 2 mL carbohydrate solution, boil. Add 5 mL dil. NH_4OH. Biol for five minutes. A rose pink colour confirms the glucose. Osazone derivative, m. p. 205ºC
Fructose	M. P. 104ºC	Sub + 2 mL ammonium molybdate solution, blue colour developed. Sub + equal amount of resorcinol+ 1 mL Conc. HCl, warm, red colour produced. *(Selivernoft test)* Osazone derivative, m. p. 205ºC
Mannose	M. P. 132ºC	Osazone derivative, m. p. 205ºC
Lactose	M. P. 203ºC	Osazone derivative, m. p. 200ºC
Sucrose	M. P. 185ºC	Osazone derivative, m. p. 205ºC

Naphthalene	M. P. 80°C	Sub + conc. H_2SO_4 + conc. HNO_3, warm, cool, pour into cold water. Yellow oil or solid compound formed. Picrate derivative, m. p. 205°C 1-nitro derivative, m.p., 61°C
Anthracene	B. P. 216°C	Saturated solution of anthracene in xylene and keep it in sunlight for few minutes. Crystals dianthracene appears. Dibromo derivatives; m.p. 221°C Picrate derivative, m. p. 138°C
Methyl alcohol	B. P. 65°C	3,5-dinitrobenzoate derivative; m.p. 109°C
Ethyl alcohol	B. P. 78°C	Iodoform test: Sub + few drops of iodine solution + NaOH solution, warm, brown colour of iodine disappears and a yellow precipitate of iodoform is formed with characteristic smell. 3,5-dinitrobenzoate derivative; m.p. 94°C
Benzyl alcohol	B. P. 205°C	3,5-dinitrobenzoate derivative; m.p. 113°C p-nitrobenzoate derivative; m.p. 86°C
Methyl salicylate	B. P. 223°C	Produce violet colour with $FeCl_3$.
Ethyl acetate	B. P. 56°C	

Group II: Compounds containing C, H, (O) and N elements			
Group II: A - Tests for acids: Compounds may contain carboxylic acid (-COOH) functional group			
S.No	**Test**	**Observation**	**Inference**
1.	Sub + saturated $NaHCO_3$ solution	Effervesces of CO_2	Amino Carboxylic acid like anthranilic acid
2.	Aqueous or alcoholic solution of sample + Few drops of $FeCl_3$	A red colour develops	Amino acid present
3.	Aqueous solution of sample + Few drops of Ninhydrin reagent	A blue colour is produced	Amino acid present
Test for Nitro acids			
1.	**Test for Nitro group** **Neutral reduction** 3 mL alcohol + Sub + 6 drops of $CaCl_2$ heat till vigorous boiling and filter into 2 mL Tollen's reagent.	Black or gray ppt.	Nitro group may present.
2.	**Acidic reduction** Sub + 2 mL conc. HCl and a pinch of Zn dust. Boil for 2 min, cool, filter and add few drops of $NaNO_2$ sol + few drops of β-naphthol in NaOH	Orange Dye stuff	Nitro group may present

Examples of Amino carboxylic acid, Amino acids, Nitro acids with literature M.P. / B. P.		
Name	**Physical Constant**	**Confirmatory test/ Derivatives**
Anthranilic acid	M. P. 144 °C	a. Sub + equal amount of $CaCl_2$, heat. Dissolve it in 2 mL of alcohol. Red colour changes violet fluorescence on standing b. Anilide derivatives m. p. 131°C

Glycine	M. P. 232 °C	a. Sub + Water + $CuSO_4$, gives blue colour. b. Sub + water + $FeCl_3$, gives red colour.
o-Nitro Benzoic Acid	M. P. 147 °C	a. Sub + soda lime, heat, nitrobenzene forms (odour of bitter almond
m-Nitro Benzoic Acid	M. P. 141°C	a. Sub + soda lime, heat, nitrobenzene forms (odour of bitter almond

Group II: B - Tests for Phenols: Compounds may contain phenolic (-OH) functional group			
S.No	**Test**	**Observation**	**Inference**
1.	Sub + 2 mL Water + 2 mL 10% NaOH	An intense yellow or orange red colour	Nitrophenols may present
	Name	**Physical Constant**	Confirmatory test/ Derivatives
1.	*o*-Nitrophenols	M. P. 45°C B. P. 216°C	a. Sub + bromine in acetic acid gives 4,6-dibromo derivatives m.p. 117°C. b. Acetate derivative, m.p. 41°C c. Benzoate derivative, m.p. 59°C d. p-Nitro benzoate derivative m.p. 141°C
2.	*m*-Nitrophenols	M. P. 97°C	a. Acetate derivative, m.p. 56°C b. Benzoate derivative, m.p. 95°C p-Nitro benzoate derivative m.p. 174°C
3.	*p*-Nitrophenols	M. P. 114°C	a. Sub + bromine in acetic acid gives 4,6-dibromo derivatives m.p. 142°C. b. Acetate derivative, m.p. 83°C c. Benzoate derivative, m.p. 142°C d. p-Nitro benzoate derivative m.p. 159°C

Group II: C – Basic Compounds – Test for amines			
S.No	**Test**	**Observation**	**Inference**
1.	Sub + Dil. HCl	Soluble	Amines present
		Reprecipitated by adding 20% NaOH (drop by drop)	Amines confirmed
2.	**Carbylamine reaction** Sub + 3-4 drops of Chloroform + 2 mL alcoholic KOH, warm	Nauseating odour of isocyanide	Presence of primary aliphatic or aromatic amine
		No reaction	Secondary or tertiary amines.
3.	**Hinsberg test:** Sub + 2 mL NaOH, shake well add 2 drops of benzene sulphonyl chloride + 2 mL of Pyridine	Yellow colour after shaking	Primary Amine
		Orange colour	Secondary amines.
		Deep Red or purple colour	Tertiary amines.
4.	**Primary Aliphatic amines:**		

	Sub + 5 mL water + 1 mL acetone + drop of sodium nitroprusside solution.	Violet colour within two min.	Primary aliphatic amine
5.	**Azo Dye test** Sub + 2 mL HCl, cool to 0 °C. In another test tube prepare ice cold solution of sodium nitrite. Mix two solutions, add into cold sol. of β-naphthol in NaOH	Orange dye stuff	Primary aromatic amine present
6.	**Nitrous acid test** Sub + 2 mL HCl + 3 mL of water and cool in ice bath + add 2 mL ice-cold 2% $NaNO_2$ solution	White or yellow white emulsion	Secondary aromatic amine present
		Yellow or yellow green colour which turns dark greenish on addition of NaOH	Tertiary aromatic amine present

Examples of amines with literature M.P. / B. P.		
Name	**Physical Constant**	**Confirmatory test/ Derivatives**
Aniline	B. P. 183°C	a. Aniline + 2 mL of ether + 10 mL of water + 1 mL dilute solution of bleaching powder, shake well. A purple colour is produced. b. Acetyl derivatives, m. p. 114°C c. Benzoyl derivative, m. p. 163°C
Benzylamine	B. P. 185°C	a. Acetyl derivatives, m. p. 60°C b. Benzoyl derivative, m. p. 105°C c. Picrate derivative, m. p. 199°C
o-Toluidine	B. P. 200°C	a. Sub + 2 mL of ether + 10 mL of water + 1 mL dilute solution of bleaching powder, shake well. Ether layer becomes brown. b. Acetyl derivatives, m. p. 112°C c. Benzoyl derivative, m. p. 144°C d. Picrate derivative, m. p. 213°C
m-toluidine	B. P. 203°C	a. Sub + 2 mL of ether + 10 mL of water + 1 mL dilute solution of bleaching powder, shake well. Water layer become yellow brown and ether layer becomes red. b. Acetyl derivatives, m. p. 66°C c. Benzoyl derivative, m. p. 125°C a. Picrate derivative, m. p. 200°C
p-toluidine	M. P. 45°C	a. Sub + few drops of dil. HCl + 2 mL water + few drops of $FeCl_3$, pale yellow colour changes to red.

Group II: D – Neutral Compounds containing C, H, (O) & N elements			
S.No	**Test**	**Observation**	**Inference**
	Test for Amines		
1.	**Hydrolysis with alkali** Sub + 2 mL Water + 2mL 10% NaOH, warm	Evolution of ammonia *(Tested with red litmus paper, bring paper near to mouth of the test tube. Red litmus paper turns of blue.)*	Amide present

2.	**Nitrous acid Test:** Sub + dil HCl, cool + add few drops of sodium nitrite solution Boil.	Brisk effervescences due to evolution of N_2	Amide present
	Test for Anilides *Boil small amount of sub with dil. HCl for 5 minutes. Cool the solution and then perform the carbylamine and azo dye test described in Test for amines*		
1.	Carbylamine test	Smell of carbylamine	Anilide may present
2.	Azo dye test	Orange dye stuff	Anilide may present
	Test for Nitro group		
1.	**Ferrous hydroxide test** Sub + 2 mL of ferrous ammonium sulphate solution + dil H_2SO_4 + alcoholic KOH, shake well	Reddish Brown ppt.	Nitro group present
2.	**Mulliken Barker Test** Sub + 5 mL alcohol+ pinch of ammonium chloride powder and a pinch of zinc dust. Boil for 5 minutes, cool and filter directly in a test tube containing 5mL tollen's reagent	A grey or black precipitate or silver mirror is observed	Nitro group is present.
3.	**Acidic reduction**: Sub + 2 mL conc. HCl + a pinch of Zn dust. Boil for 2 min, cool, filter and add few drops of $NaNO_2$ sol + few drops of β-naphthol in NaOH	Orange Dye stuff	Nitro group may present

Examples of amides, anilides and nitro compounds with literature M.P. / B. P.		
Name	**Physical Constant**	**Confirmatory test/ Derivatives**
Urea	M. P. 132°C	a. Biuret test - Urea in test tube, heat until it melts. Dissolve in 1 mL NaOH and add dilute $CuSO_4$ solution, violet colour is produced. b. Urea + 3 mL of conc. HNO_3, boil and cool; urea nitrate (white crystals) is formed m.p. 163°C.
Benzamide	M. P. 129°C	a. Benzamide + dilute NaOH, boil for five minutes, cool and acidify with dilute HCl. A white precipitate of benzoic acid is obtained which is confirmed by Neutral test solution. m.p. 121°C
Phthalimide	M. P. 235°C	a. Phthalimide + dilute NaOH, boil for five minutes, cool and acidify with dilute HCl. A white precipitate of phthalic acid. m.p. 195°C
Acetanilide	M. P. 114°C	a. Nitration reaction: Acetanilide + conc. HNO_3 + conc. H_2SO_4, p-nitroacetanilide produced. m.p. 216°C

Nitrobenzene	B. P. 211°C	a. Acidic reduction: 1 mL of sub + a pinch of zinc dust or $SnCl_2$ + 2 mL conc. HCl, boil for 5 min. Aniline is produced and confirmed by Azo dye test and carbylamine test.

Group III Compounds containing C, H, (O), N & S elements			
Group III: A – Acidic Compounds			
S.No	**Test**	**Observation**	**Inference**
1.	**Test for amino sulphonic acid** Sub + saturated solution of $NaHCO_3$. Perform Azo- dye test	Soluble with strong effervesces. Orange dye stuff	Amino sulphonic acid present Primary aromatic amine
Group III: B – Neutral Compounds			
1.	**Test for thioureas** Sub + 2 mL NaOH solution, boil for 1 minutes, cool and add lead acetate solution.	Dark brown or black precipitate	Thiourea is present
		No colour	Sulphonamide is present
2.	**Test for sulphonamide** Sub + 2 mL Water + 2mL 10% NaOH, warm	Evolution of ammonia	Sulphonamide present.

Name	**Physical Constant**	**Confirmatory test/ Derivatives**
Thiourea	M. P. 182°C	a. Thiourea + NaOH solution, boil, NH_3 is evolved. + 5 mL water + few drops of $FeCl_3$, blood red colour is produced.

Preparation of Derivatives

Class of compound	Derivative
Carboxylic acids	Anilide, Amide, p-toluidide
Phenols	Benzoate, Acetate, Bromo-derivative, Toulene-p-sulphonate, p-nitrobenzoates, aryloxyacetic acid etc.
Alcohols	3,5-dinitrobenzoate
Amines	Benzoyl, Acetyl, Picrate
Nitro compounds	Nitro derivative
Aldehydes and ketones	Semicarbazone, 2,4-Dinitrophenyl Hydrazone, Oxime
Amides	Acid, Nitro
Carbonyl compounds	Semicarbazone, 2,4-Dinitrophenylhydrazones, Oximes
Esters	Acid (hydrolysis)
Ethers and Hydrocarbons	Nitro, Picrate
Carbohydrates	Osazone, Benzoyl
Halogen compounds	Nitro

Derivatives of carboxylic acids

Solution A: Place 0.5 – 1.0 g of carboxylic acid in a dry round bottomed flask fitted with reflux condenser add 2.5- 5.0 mL of thionyl chloride dropwise, reflux on hot water bath for about 30 mins.

Prepare either/all of following derivatives from solution A.

1. Preparation of amide: From solution A distill off the excess of thionyl chloride, cool and add ammonia drop wise till a solid precipitate is formed and cool for 5 minutes. Recrystallize the amide from hot aqueous alcohol. Record M.P.

2. Preparation of anilide: Dilute acid chloride with 5 mL of pure ether or benzene, add a solution of 2 g of aniline in 15-20 mL same solvent until odour of acid chloride has disappeared. Shake the resulting solution with excess of dilute hydrochloric acid to remove aniline and its salts. Evaporate the remaining solvent. recrystallize the anilide form dilute ethanol or toluene. Record M.P.

3. Preparation of p-toluidides: Proceed same as under anilide but substitute p-toluidine for aniline. Record M.P.

Derivatives of phenols

1. Preparation of benzoate (Schötten-Baumann method) derivative: To 0.5 g of compound add 10 mL of 10% sodium hydroxide in a well-corked boiling tube or a small conical flask. Add 2 mL of benzoyl chloride dropwise and shake the mixture vigorously with occasional cooling under the tap or in ice-water. After 15 min the solid benzoate separates out [the solution should be alkaline at the end of the reaction; if not alkaline, or if the product is oily, add a solid pellet of sodium hydroxide and shake again.] collect the benzoate, wash thoroughly with cold water, and recrystallize from alcohol. Record M.P.

2. Preparation of acetate derivative: To 0.5 g of compound add 10 mL of 10% sodium hydroxide and an equal quantity of crushed ice, followed by 2 mL acetic anhydride. Shake the mixture vigorously in a stoppered test tube until the acetate separates. Filter the product and recrystallize from alcohol. Record M.P.

3. Preparation of bromo derivatives: Dissolve 0.5 g of compound in 5 mL dilute acetic acid then, add solution of 3-5 mL bromine dissolved in 10-15 mL acetic acid dropwise until the colour of bromine persists. Allow the mixture to stand for 10-15 minutes, pour in a crushed ice with stirring, filter the product separated, wash and recrystallize from alcohol. Record M.P.

Derivatives of alcohols

1. Preparation of 3,5-dinitrobenzoate derivative: Mix 0.6 mL of the alcohol with about 0.2 g of 3,5-dinitrobenzoyl chloride in around-bottom flask fitted to a reflux condenser. Reflux the mixture for about 15 - 30 min., Cool the solution and add about 5 – 10 mL of dilute sodium bicarbonate solution to neutralize remaining 3,5-dinitrobenzoyl chloride and hydrochloric acid generated from reaction. Cool this solution in an ice-water bath, and collect the crude crystalline product. Recrystallize the product from aqueous ethanol. Record M.P.

Derivatives of amines

1. Preparation of acetyl derivatives (acetamides): Reflux gently in a small test tube under a short air condenser 1 g of amine with 3 mL acetic anhydride for 15 min. Cool the reaction mixture and pour into 20 mL cold water. Boil to decompose the excess acetic anhydride. Cool and filter by suction the insoluble derivative. Recrystallize from ethanol. Record M.P.

2. Preparation of benzoyl derivatives (benzamides): Suspend 1 g of the amine in 10 - 15 mL of 10% aqueous sodium hydroxide in a well-corked flask, and add dropwise 2 mL benzoyl chloride, with constant shaking. Shake vigorously for 5-10 min until the odour of the benzoyl chloride has disappeared. Ensure that the mixture remains alkaline. Filter off the solid derivative, wash with a little cold water and recrystallize from ethanol. Record M.P.

3. Preparation of picrate Derivative: Dissolve 0.5 g or 1 mL of the amine in 5 mL ethanol and add 3-4 mL cold alcoholic saturated picric acid solution. Warm on a water bath for 5 min; allow to cool, picrate separates out as bulky solid, filter wash with cold water. Recrystallize from ethanol. Record M.P.

Derivative of nitro compounds

1. Nitration: Take 2 mL Conc. HNO_3 and 2.5 mL Conc. H_2SO_4 in RB flask fitted with condenser. Then add 1.5 mL of nitrobenzene. Heat the reaction mixture with frequent shaking in water bath for an hour. Pour the contents of the flask while hot in 100 mL ice cold water in a beaker. Filter the product wash with cold water and recrystallize from alcohol. Record M.P.

Derivatives of amides

1. Hydrolysis (Preparation of acid): Place 1 g of compound in RB flask fitted with reflux condenser. Add 15 mL of 10% aqueous NaOH solution and reflux for 20-25 minutes. Stop heating, cool the contents and acidify with dil. hydrochloric acid solution till it is acidic to litmus. Carboxylic acid separates out as solid, filter recrystallize from water.

2. Nitration: Take 2 mL conc. HNO_3 and 2.5 mL Conc. H_2SO_4 in RB flask fitted with condenser. Then add 1 g of nitrobenzene. Heat the reaction mixture with stirring in water bath for 30 minutes. Pour the

contents of the flask while hot in 100 mL ice cold water in a beaker. Filter the product wash with cold water. Record M.P.

Derivatives of aldehydes and ketones or carbonyl compounds

1. Semicarbazones: Dissolve 1 g semicarbazide hydrochloride and 1.5 g sodium acetate in 8 – 10 mL water, add the 0.3 mL aldehyde or ketone. Shake the mixture for a few minutes and then cool in ice water. Filter off the crystals, wash with a little cold water and recrystallise from methanol or ethanol. Record M.P.

2. 2,4-Dinitrophenylhydrazones: Suspend 0.25 g of 2,4-dinitrophenylhydrazine in 5 mL of methanol and add 0.5 mL of conc. sulphuric acid cautiously. Filter the warm solution and add a solution of 0.2 g of the carbonyl compound in 1 mL of methanol. Recrystallise the derivative from methanol, ethanol or ethyl acetate. Record M.P.

3. Oximes: Dissolve 0.5 g Hydroxylamine hydrochloride 2 - 5 mL in water. Add 2 mL 10% sodium hydroxide and the 0.2 - 0.3 g carbonyl compound dissolved in 1 - 2 mL ethanol, the mixture warmed on a steam bath for 10 min and then, cooled in ice. Crystallize by scratching the sides of the test tube with a glass rod. Recrystallise the derivative from ethanol. Record M.P.

Derivatives of carbohydrates

1. Osazone derivative: Take 5 mL of the sugar solution in a test tube, add 0.5 g of phenyl hydrazine reagent, add 0.1 g of sodium acetate and a few drops of glacial acetic acid. The contents are mixed well and placed in a boiling water bath. Cool the solution to room temperature and transfer a few crystals onto a glass slide, cover it with a cover slip and observe the shape of the crystals under a light microscope. Needle shaped yellow crystals of fructosazone are formed within 5 – 7 minutes indicates presence of fructose, Needle shaped yellow crystals of glucosazone are formed within 10 minutes indicates presence of glucose, Sunflower shaped crystals of maltosazone are formed in 30 minutes indicates presence of maltose, Cotton ball shaped crystals of lactosazone formed, indicates presence of lactose.

2. Benzoylation: Dissolve 1 g of carbohydrate in in 10 of 20 % aqueous NaOH solution and add 2mL of benzoyl chloride. Cork the flask and shake vigorously for 10 minutes where by a white solid of penta benzoyl derivative separates out. Filter and dry it. Record M.P.

Derivatives of ethers and hydrocarbons

1. Nitro derivative: To 1 mL or 0.5 g of compound add a mixture of 2 mL of conc. HNO_3 and 3 mL of conc.H_2SO_4 drop-wise. Heat the reaction mixture on water bath for 30 minute and transfer the contents in beaker containing ice cold water. Filter the derivative separated and dry it. [Nitration for some compound is also take place at RT]. Record M.P.

2. Picrate Derivative: Mix about 0.5 g or 1 mL of the compound dissolved in 2-3 mL ethanol with 3 - 4 mL alcoholic saturated picric acid solution. Heat the reaction on water bath for 15 minutes and stir well picrate separates out as bulky solid, filter wash with cold water. Recrystallize from ethanol. Record M.P.

Experiment No. 23
Systematic Qualitative Analysis – Compound (C-1)

1. Aim
To identify the given unknown organic compound C-1 by Systematic Qualitative Analysis.

2. Practical Significance
Qualitative analysis is the analysis of the functional group present in a given organic compound. For example, if a compound is taken, the qualitative analysis would be more focused on finding the elements and the functional group present in the compound rather than study as to how much they are present. The identification and analysis of unknown organic compounds make a very important aspect of experimental organic chemistry. A systematic approach based on the scheme helps in fetching good results as there is no definite set of procedures which can be applied for all.

3. Practical Outcomes
After completion of this practical, the students will be able to:

PrO	Practical Outcomes	Mapped CO	BTL
1	Explain the principle and reaction involved in specific test.	CO 5	2
2	Write systematic analytical reports.	CO 5	3
3	Identify the given unknown organic compound through systematic qualitative analysis.	CO 5	5
4	Follow cleanliness, safety and ethical practices.	CO 5	5
5	Demonstrate working as a leader or team member.	CO 5	5

4. Systematic qualitative analysis of organic compounds
The systematic qualitative analysis of organic compounds includes the following different steps.

a. Preliminary Tests
b. Determination of Physical Constant
c. Detection of elements
d. Detection of Functional Group
e. Identification of the compound/drug by search of literature with similar physical and chemical properties.
f. Confirmative Test (Specific colour reaction or preparing suitable derivative.)

5. Requirements
a. Glasswares: Test tubes, Beakers, Measuring cylinder, Graduated pipettes, Evaporating dish, Water bath, Thiele's tube, Thermometer, Wire gauze, Tripod stand.
b. Chemicals: All general and table reagents.

6. Requirements used
_____

_____

7. Procedure
a. Perform stepwise Systematic Qualitative Analysis of unknown organic compounds.
b. Refer chart given in “Systematic Qualitative Analysis” for the sequence of various tests.

8. Observations

A. Preliminary Tests:

Sr. No.	Test	Observation	Inference
1	Physical State		
2	Colour		
3	Odour		
4	**Solubility Test** Sub + Water, warm if necessary		
A	**Soluble in water** Litmus Paper test **Blue Litmus Paper Test** **If acidic,** add substance to 10% Sodium Bicarbonate solution. **If non acidic –** perform a red Litmus Paper Test		
B	**Insoluble in water** Sub + 10% $NaHCO_3$. Sub + 10% NaOH Sub + Dil HCl		
5	**Action of Reagents**		
A.	**Action of cold NaOH** Sub + 2 mL Water + 2mL 10% NaOH		
B.	**Action of Hot NaOH** Warm the above mixture strongly		
C.	**Action of Hot Conc. H_2SO4** Sub + 1 mL conc. H_2SO_4, warm		
D.	**Action of Na_2CO_3 Solution** Sub + 5 mL 10%. Na_2CO_3 solution		
E.	**Test for unsaturation** 1. Baeyer's Test 2. Bromine water Test.		
F.	**Action of Ferric Chloride solution** Sub + 2 mL water + 2 mL $FeCl_3$ solution, shake well		
G.	**Heating on Copper Gauze** Small copper foil and heat it in the flame. Place 0.2 g sample on it and heat in the flame.		
H.	**Heating with Soda Lime** Sub + 2g finely powdered soda lime + 1 g coarse soda lime and heat		

Conclusion: On the basis of the tests performed above the given organic compound is

a. Aromatic / Aliphatic
b. Saturated / Unsaturated
b. Acid / Phenol/ Base/ Neutral

B. Determination of Physical Constant:

Conclusion: The melting / boiling point of a given organic compound was found to be _____.

C. Detection of Elements: Sodium Fusion Test / Lessaigne's Test

Sr. No.	Test	Observation	Inference
1	**Test for Nitrogen**		
2	**Test for Sulphur**		
3	**Test for nitrogen and sulphur together**		
4	**Test for Halogens**		

Conclusion: The given organic compound found to contain _____ elements.

D. Detection of functional group / groups for compounds containing _____ elements and acidic/basic/neutral/phenolic in nature.

Sr. No.	Test	Observation	Inference

Conclusion: The given organic compound was found to contain_____ functional group / groups.

E. Confirmatory Tests: Specific colour reaction or derivative preparation

Sr. No.	Test	Observation	Inference

9. Result

The systematic qualitative analysis of compound C1 shows that, it is

a) State : _____

b) Aromatic / Aliphatic : _____

c) Soluble in : _____

d) Elements Present : _____

e) Functional Group : _____

f) Physical Constant : _____

g) Name of the Compound : _____

h) Derivate if any with M.P.: _____

i) Draw chemical structure of compound from official book.

10. Conclusion

Systematic qualitative analysis of a given organic compound was performed.

11. References

a. Indian Pharmacopoeia 2022.
b. Vogel's textbook of practical organic chemistry, Fifth edition, Pearson education.

12. Practical Related Questions

a. Write name & procedure of preliminary tests which can be performed for finding out unsaturation in the organic compound.
b. How will you identify the presence of carboxylic acid (-COOH) functional group?
c. Why does salicylic acid give violet colour with ferric chloride solution?
d. All aromatic compounds give a sooty flame on burning. Why?
e. Identify the nature of compound which is acidic to litmus but does not give effervescence with sodium bicarbonate.

(*Space for Answers*)

MAHARASHTRA STATE BOARD OF TECHNICAL EDUCATION MUMBAI

13. Assessment Scheme

Particular	Understanding the basic concept (Intellectual skill)	Activities (Motor skill)	Cleanliness & Handling (Affective domain)	Viva-voce / Answers Written	Total	Signature of teacher
Marks Obtained						
Max Marks	**02**	**05**	**01**	**02**	**10**	

Experiment No. 24
Systematic Qualitative Analysis – Compound (C-2)

1. Aim
To identify the given unknown organic compound C-2 by Systematic Qualitative Analysis.

2. Practical Significance
Qualitative analysis is the analysis of the functional group present in a given organic compound. For example, if a compound is taken, the qualitative analysis would be more focused on finding the elements and the functional group present in the compound rather than study as to how much they are present. The identification and analysis of unknown organic compounds make a very important aspect of experimental organic chemistry. A systematic approach based on the scheme helps in fetching good results as there is no definite set of procedures which can be applied for all.

3. Practical Outcomes
After completion of this practical, the students will be able to:

PrO	Practical Outcomes	Mapped CO	BTL
1	Explain the principle and reaction involved in specific test.	CO 5	2
2	Write systematic analytical reports.	CO 5	3
3	Identify the given unknown organic compound through systematic qualitative analysis.	CO 5	5
4	Follow cleanliness, safety and ethical practices.	CO 5	5
5	Demonstrate working as a leader or team member.	CO 5	5

4. Systematic qualitative analysis of organic compounds
The systematic qualitative analysis of organic compounds includes the following different steps.

a. Preliminary Tests
b. Determination of Physical Constant
c. Detection of elements
d. Detection of Functional Group
e. Identification of the compound/drug by search of literature with similar physical and chemical properties.
f. Confirmative Test (Specific colour reaction or preparing suitable derivative.)

5. Requirements
a. Glasswares: Test tubes, Beakers, Measuring cylinder, Graduated pipettes, Evaporating dish, Water bath, Thiele's tube, Thermometer, Wire gauze, Tripod stand.
b. Chemicals: All general and table reagents.

6. Requirements used
_____

_____

7. Procedure
a. Perform stepwise Systematic Qualitative Analysis of unknown organic compounds.
b. Refer chart given in "Systematic Qualitative Analysis" for the sequence of various tests.

8. Observations

A. Preliminary Tests:

Sr. No.	Test	Observation	Inference
1	Physical State		
2	Colour		
3	Odour		
4	**Solubility Test** Sub + Water, warm if necessary		
A	**Soluble in water** Litmus Paper test **Blue Litmus Paper Test** **If acidic,** add substance to 10% Sodium Bicarbonate solution. **If non acidic –** perform a red Litmus Paper Test		
B	**Insoluble in water** Sub + 10% $NaHCO_3$. Sub + 10% NaOH Sub + Dil HCl		
5	**Action of Reagents**		
A.	**Action of cold NaOH** Sub + 2 mL Water + 2mL 10% NaOH		
B.	**Action of Hot NaOH** Warm the above mixture strongly		
C.	**Action of Hot Conc. H_2SO4** Sub + 1 mL conc. H_2SO_4, warm		
D.	**Action of Na_2CO_3 Solution** Sub + 5 mL 10%. Na_2CO_3 solution		
E.	**Test for unsaturation** 1. Baeyer's Test 2. Bromine water Test.		
F.	**Action of Ferric Chloride solution** Sub + 2 mL water + 2 mL $FeCl_3$ solution, shake well		
G.	**Heating on Copper Gauze** Small copper foil and heat it in the flame. Place 0.2 g sample on it and heat in the flame.		
H.	**Heating with Soda Lime** Sub + 2g finely powdered soda lime + 1 g coarse soda lime and heat		

Conclusion: On the basis of the tests performed above the given organic compound is

a. Aromatic / Aliphatic
b. Saturated / Unsaturated
c. Acid / Phenol/ Base/ Neutral

B. Determination of Physical Constant:

Conclusion: The melting / boiling point of a given organic compound was found to be _____.

C. Detection of Elements: Sodium Fusion Test / Lessaigne's Test

Sr. No.	Test	Observation	Inference
1	**Test for Nitrogen**		
2	**Test for Sulphur**		
3	**Test for nitrogen and sulphur together**		
4	**Test for Halogens**		

Conclusion: The given organic compound found to contain _____ elements.

D. Detection of functional group / groups for compounds containing _____ elements and acidic/basic/neutral/phenolic in nature.

Sr. No.	Test	Observation	Inference

Conclusion: The given organic compound was found to contain_____ functional group / groups.

E. Confirmatory Tests: Specific colour reaction or derivative preparation

Sr. No.	Test	Observation	Inference

9. Result

The systematic qualitative analysis of compound C1 shows that, it is

a) State : _____

b) Aromatic / Aliphatic : _____

c) Soluble in : _____

d) Elements Present : _____

e) Functional Group : _____

f) Physical Constant : _____

g) Name of the Compound : _____

h) Derivate if any with M.P.: _____

i) Draw chemical structure of compound from official book.

10. Conclusion

Systematic qualitative analysis of a given organic compound was performed.

11. References

a. Indian Pharmacopoeia 2022.
b. Vogel's textbook of practical organic chemistry, Fifth edition, Pearson education.

12. Practical Related Questions

a. Explain the basic principle of Lassaigne's test.
b. Write reaction involved in nitrogen and sulphur test.
c. Write procedure for preparation of sodium fusion extract.
d. How acetic acid is differed from glacial acetic acid.
e. Give detection test for nitrogen and sulphur together in elemental analysis?

(*Space for Answers*)

MAHARASHTRA STATE BOARD OF TECHNICAL EDUCATION MUMBAI

13. Assessment Scheme

Particular	Understanding the basic concept (Intellectual skill)	Activities (Motor skill)	Cleanliness & Handling (Affective domain)	Viva-voce / Answers Written	Total	Signature of teacher
Marks Obtained						
Max Marks	**02**	**05**	**01**	**02**	**10**	

Experiment No. 25
Systematic Qualitative Analysis – Compound (C-3)

1. Aim
To identify the given unknown organic compound C-3 by Systematic Qualitative Analysis.

2. Practical Significance
Qualitative analysis is the analysis of the functional group present in a given organic compound. For example, if a compound is taken, the qualitative analysis would be more focused on finding the elements and the functional group present in the compound rather than study as to how much they are present. The identification and analysis of unknown organic compounds make a very important aspect of experimental organic chemistry. A systematic approach based on the scheme helps in fetching good results as there is no definite set of procedures which can be applied for all.

3. Practical Outcomes
After completion of this practical, the students will be able to:

PrO	Practical Outcomes	Mapped CO	BTL
1	Explain the principle and reaction involved in specific test.	CO 5	2
2	Write systematic analytical reports.	CO 5	3
3	Identify the given unknown organic compound through systematic qualitative analysis.	CO 5	5
4	Follow cleanliness, safety and ethical practices.	CO 5	5
5	Demonstrate working as a leader or team member.	CO 5	5

4. Systematic qualitative analysis of organic compounds
The systematic qualitative analysis of organic compounds includes the following different steps.

a. Preliminary Tests
b. Determination of Physical Constant
c. Detection of elements
d. Detection of Functional Group
e. Identification of the compound/drug by search of literature with similar physical and chemical properties.
f. Confirmative Test (Specific colour reaction or preparing suitable derivative.)

5. Requirements
a. Glasswares: Test tubes, Beakers, Measuring cylinder, Graduated pipettes, Evaporating dish, Water bath, Thiele's tube, Thermometer, Wire gauze, Tripod stand.

b. Chemicals: All general and table reagents.

6. Requirements used
_____

_____

7. Procedure
a. Perform stepwise Systematic Qualitative Analysis of unknown organic compounds.
b. Refer chart given in "Systematic Qualitative Analysis" for the sequence of various tests.

8. Observations

A. Preliminary Tests:

Sr. No.	Test	Observation	Inference
1	Physical State		
2	Colour		
3	Odour		
4	**Solubility Test** Sub + Water, warm if necessary		
A	**Soluble in water** Litmus Paper test **Blue Litmus Paper Test** **If acidic,** add substance to 10% Sodium Bicarbonate solution. **If non acidic –** perform a red Litmus Paper Test		
B	**Insoluble in water** Sub + 10% $NaHCO_3$. Sub + 10% NaOH Sub + Dil HCl		
5	**Action of Reagents**		
A.	**Action of cold NaOH** Sub + 2 mL Water + 2mL 10% NaOH		
B.	**Action of Hot NaOH** Warm the above mixture strongly		
C.	**Action of Hot Conc. H_2SO4** Sub + 1 mL conc. H_2SO_4, warm		
D.	**Action of Na_2CO_3 Solution** Sub + 5 mL 10%. Na_2CO_3 solution		
E.	**Test for unsaturation** 1. Baeyer's Test 2. Bromine water Test.		
F.	**Action of Ferric Chloride solution** Sub + 2 mL water + 2 mL $FeCl_3$ solution, shake well		
G.	**Heating on Copper Gauze** Small copper foil and heat it in the flame. Place 0.2 g sample on it and heat in the flame.		
H.	**Heating with Soda Lime** Sub + 2g finely powdered soda lime + 1 g coarse soda lime and heat		

Conclusion: On the basis of the tests performed above the given organic compound is

a. Aromatic / Aliphatic
b. Saturated / Unsaturated
c. Acid / Phenol/ Base/ Neutral

B. Determination of Physical Constant:

Conclusion: The melting / boiling point of a given organic compound was found to be _____.

C. Detection of Elements: Sodium Fusion Test / Lessaigne's Test

Sr. No.	Test	Observation	Inference
1	**Test for Nitrogen**		
2	**Test for Sulphur**		
3	**Test for nitrogen and sulphur together**		
4	**Test for Halogens**		

Conclusion: The given organic compound found to contain _____ elements.

D. Detection of functional group / groups for compounds containing _____ elements and acidic/basic/neutral/phenolic in nature.

Sr. No.	Test	Observation	Inference

Conclusion: The given organic compound was found to contain_____ functional group / groups.

E. Confirmatory Tests: Specific colour reaction or derivative preparation

Sr. No.	Test	Observation	Inference

9. Result

The systematic qualitative analysis of compound C1 shows that, it is

a) State : _____

b) Aromatic / Aliphatic : _____

c) Soluble in : _____

d) Elements Present : _____

e) Functional Group : _____

f) Physical Constant : _____

g) Name of the Compound : _____

h) Derivate if any with M.P.: _____

i) Draw chemical structure of compound from official book.

10. Conclusion

Systematic qualitative analysis of a given organic compound was performed.

11. References

a. Indian Pharmacopoeia 2022.
b. Vogel's textbook of practical organic chemistry, Fifth edition, Pearson education.

12. Practical Related Questions

a. Write principle involved in Diazotization & Phthalein test.
b. Write the name and procedure of a test functional group present in compound analyzed.
c. What are derivatives/ name the possible derivatives, which can be prepared from the compound analyzed.
d. Write chemical reactions involved in the preparation of derivatives.

(*Space for Answers*)

MAHARASHTRA STATE BOARD OF TECHNICAL EDUCATION MUMBAI

13. Assessment Scheme

Particular	Understanding the basic concept (Intellectual skill)	Activities (Motor skill)	Cleanliness & Handling (Affective domain)	Viva-voce / Answers Written	Total	Signature of teacher
Marks Obtained						
Max Marks	**02**	**05**	**01**	**02**	**10**	

Experiment No. 26
Systematic Qualitative Analysis – Compound (C-4)

1. Aim
To identify the given unknown organic compound C-4 by Systematic Qualitative Analysis.

2. Practical Significance
Qualitative analysis is the analysis of the functional group present in a given organic compound. For example, if a compound is taken, the qualitative analysis would be more focused on finding the elements and the functional group present in the compound rather than study as to how much they are present. The identification and analysis of unknown organic compounds make a very important aspect of experimental organic chemistry. A systematic approach based on the scheme helps in fetching good results as there is no definite set of procedures which can be applied for all.

3. Practical Outcomes
After completion of this practical, the students will be able to:

PrO	Practical Outcomes	Mapped CO	BTL
1	Explain the principle and reaction involved in specific test.	CO 5	2
2	Write systematic analytical reports.	CO 5	3
3	Identify the given unknown organic compound through systematic qualitative analysis.	CO 5	5
4	Follow cleanliness, safety and ethical practices.	CO 5	5
5	Demonstrate working as a leader or team member.	CO 5	5

4. Systematic qualitative analysis of organic compounds
The systematic qualitative analysis of organic compounds includes the following different steps.

a. Preliminary Tests
b. Determination of Physical Constant
c. Detection of elements
d. Detection of Functional Group
e. Identification of the compound/drug by search of literature with similar physical and chemical properties.
f. Confirmative Test (Specific colour reaction or preparing suitable derivative.)

5. Requirements
a. Glasswares: Test tubes, Beakers, Measuring cylinder, Graduated pipettes, Evaporating dish, Water bath, Thiele's tube, Thermometer, Wire gauze, Tripod stand.
b. Chemicals: All general and table reagents.

6. Requirements used
_____

_____

7. Procedure
a. Perform stepwise Systematic Qualitative Analysis of unknown organic compounds.
b. Refer chart given in "Systematic Qualitative Analysis" for the sequence of various tests.

8. Observations

A. Preliminary Tests:

Sr. No.	Test	Observation	Inference
1	Physical State		
2	Colour		
3	Odour		
4	**Solubility Test** Sub + Water, warm if necessary		
A	**Soluble in water** Litmus Paper test **Blue Litmus Paper Test** **If acidic,** add substance to 10% Sodium Bicarbonate solution. **If non acidic –** perform a red Litmus Paper Test		
B	**Insoluble in water** Sub + 10% $NaHCO_3$. Sub + 10% NaOH Sub + Dil HCl		
5	**Action of Reagents**		
A.	**Action of cold NaOH** Sub + 2 mL Water + 2mL 10% NaOH		
B.	**Action of Hot NaOH** Warm the above mixture strongly		
C.	**Action of Hot Conc. H_2SO4** Sub + 1 mL conc. H_2SO_4, warm		
D.	**Action of Na_2CO_3 Solution** Sub + 5 mL 10%. Na_2CO_3 solution		
E.	**Test for unsaturation** 1. Baeyer's Test 2. Bromine water Test.		
F.	**Action of Ferric Chloride solution** Sub + 2 mL water + 2 mL $FeCl_3$ solution, shake well		
G.	**Heating on Copper Gauze** Small copper foil and heat it in the flame. Place 0.2 g sample on it and heat in the flame.		
H.	**Heating with Soda Lime** Sub + 2g finely powdered soda lime + 1 g coarse soda lime and heat		

Conclusion: On the basis of the tests performed above the given organic compound is

a. Aromatic / Aliphatic
b. Saturated / Unsaturated
c. Acid / Phenol/ Base/ Neutral

B. Determination of Physical Constant:

Conclusion: The melting / boiling point of a given organic compound was found to be _____.

C. Detection of Elements: Sodium Fusion Test / Lessaigne's Test

Sr. No.	Test	Observation	Inference
1	**Test for Nitrogen**		
2	**Test for Sulphur**		
3	**Test for nitrogen and sulphur together**		
4	**Test for Halogens**		

Conclusion: The given organic compound found to contain _____ elements.

D. Detection of functional group / groups for compounds containing _____ elements and acidic/basic/neutral/phenolic in nature.

Sr. No.	Test	Observation	Inference

Conclusion: The given organic compound was found to contain_____ functional group / groups.

E. Confirmatory Tests: Specific colour reaction or derivative preparation

Sr. No.	Test	Observation	Inference

9. Result

The systematic qualitative analysis of compound C1 shows that, it is

a) State : _____

b) Aromatic / Aliphatic : _____

c) Soluble in : _____

d) Elements Present : _____

e) Functional Group : _____

f) Physical Constant : _____

g) Name of the Compound : _____

h) Derivate if any with M.P.: _____

i) Draw chemical structure of compound from official book.

10. Conclusion

Systematic qualitative analysis of a given organic compound was performed.

11. References

a. Indian Pharmacopoeia 2022.
b. Vogel's textbook of practical organic chemistry, Fifth edition, Pearson education.

12. Practical Related Questions

a. Write chemical reactions involved in the preparation of derivatives.
b. How will you detect an amine ($-NH_2$) functional group?
c. Write the difference between amines and amides?
d. What are amines? Write different types of amines with examples.
e. Write the chemical reaction involved in azo dye test.

(*Space for Answers*)

MAHARASHTRA STATE BOARD OF TECHNICAL EDUCATION MUMBAI

13. Assessment Scheme

Particular	Understanding the basic concept (Intellectual skill)	Activities (Motor skill)	Cleanliness & Handling (Affective domain)	Viva-voce / Answers Written	Total	Signature of teacher
Marks Obtained						
Max Marks	**02**	**05**	**01**	**02**	**10**	

Maharashtra State Board of Technical Education, Mumbai.

Guidelines to Conduct Sessional Practical Examination

Course Name & Abbr: Pharmaceutical Chemistry-Practical (PCP)
Course Code: 20052 **Year**: First Year (PH1J)
Max Time: 3 hrs **Max. Marks**: 80

Q. 1. Synopsis **10 M**
(5 questions of 2 marks each based on may be asked as per the sessional syllabus.)

Q. 2. Experiments **50 M**

a. Major experiment 30 M
To identify the given unknown organic compound by Systematic Qualitative Analysis.
OR
To perform assay of _____ as per IP.

b. Minor experiment 20 M
To perform and report the limit tests for _____ on the given samples as per IP.
OR
To perform and report the identification test for the cations or anions on the given sample.
OR
To prepare and standardize _____ solution as per IP. (*If assay is not asked in the major experiment, preparation and standardization may be asked.*)
OR
To perform and report identification test on the given sample of _____ as per IP.

Q.3. Viva voce **10 M**
(Viva should be conducted on practical and theory-based questions)

O.4. Practical Record Maintenance **10 M**

Maharashtra State Board of Technical Education, Mumbai.

Guidelines to Conduct Annual Practical Examination

Course Name & Abbr: Pharmaceutical Chemistry-Practical (PCP)
Course Code: 20052 **Year**: First Year (PH1J)
Max Time: 3 hrs **Max. Marks**: 80

Q. 1. Synopsis **10 M**

(5 questions of 2 marks each based on limit test, volumetric analysis, identification tests, synthesis of organic compound, systematic qualitative analysis may be asked.)

Q. 2. Experiments **60 M**

a. Major experiment 40 M

To identify the given unknown organic compound by Systematic Qualitative Analysis.

OR

To perform assay of _____ as per IP.

b. Minor experiment 20 M

To perform and report the limit tests for _____ on the given samples as per IP.

OR

To perform and report the identification test for the cations or anions on the given sample.

OR

To prepare and standardize _____ solution as per IP. (*If assay is not asked in the major experiment, preparation and standardization may be asked.*)

OR

To perform and report identification test on the given sample of _____ as per IP.

Q.3. Viva voce **10 M**

(Viva should be conducted on practical and theory-based questions)