CHEMISTRY LABORATORY MANUAL

CMR INSTITUTE OF TECHNOLOGY
DEPARTMENT OF CHEMISTRY

CMRIT/CHEL/Man/2023-24

General instructions

- Keep your working table clean and tidy.
- Keep your glassware/apparatus clean.
- Keep the reagents in their proper place after use. Do not alter their order. Do not contaminate the reagents.
- Turn offgas valve and water taps after use.
- Do not throw any waste paper into the sink. Throw them into the trash bin provided.
- Do not pour concentrated acids into the sink.
- Make note of the observations during experiment and not after completion. Have a notebook for recording and keep it away from the reagents and sink.
- Use specified quantities of reagents.
- Do not walk bare footed in the laboratory.
- Mix solutions well after addition of every reagent.
- Do not use wet hand during weighing.

During and after experimental data collection:

- Please collect experimental data with extreme caution and patience. Try your best to avoid all kinds of errors that might creep in and cause deviations in your data. If you collect the data properly correct results are always assured.
- After collection of experimental data, think whether it makes sense to you and if it is reasonable. Understand all the equations and calculations. Access critically if the results are reasonable and do they make any sense at all.

Dos and don'ts, ethics, and safety rules in the chemistry lab:

- Do not make up/fabricate the results. No fake results must be produced.
- Pay full attention to the experiment you are doing and maintain silence.
- Ask your laboratory instructor if you have questions and do not talk with others if not required.
- Do not copy experimental data and results from others or previous years record book.
- Do not drink or eat food in the lab.
- Handle chemicals with proper care and ask your lab instructor if you have any questions related to proper handling and disposal of chemicals.
- Operate gas burners carefully and be cautious while dealing with the fire.
- Do not sabotage others experiments.

Observations:

Observations are important physical phenomena occurring within specified experimental conditions. And you must state only such happenings that take place during the course of experiment. Observations must not change for a particular experiment but there is a possibility of addition of new observations that were overlooked before. Example: Say you slowly add solution of sodium hydroxide (NaOH) to the water containing phenolphthalein indicator. The basicity of water increases with addition of NaOH solution and that leads to change in the color of solution from colorless to pink. *So the observation you made here is the "change in the color of the solution from colorless to pink".*

PRACTICAL MODULE (CSE)

PRACTICAL MODULE (ECE)

Experiment A1(CSE): Chemical Structure drawing using software: ChemDraw

About the software:

ChemDraw is a chemical drawing and visualization software developed by PerkinElmer. It is widely used by chemists and researchers for drawing chemical structures and reactions, designing molecules, and creating professional-looking scientific illustrations.

Step-by-step guide on how to draw a chemical structure using ChemDraw software:

1. Open ChemDraw software and create a new document.

2. Choose the type of bond that you want to draw (single bond, double bond, triple bond, etc.) by selecting it from the toolbar.

3. Draw the atoms in the chemical structure by clicking on the appropriate element symbol in the toolbar, then clicking on the canvas where you want to place it. You can also type the element symbol directly on the canvas to add it.

4. Add bonds between the atoms by selecting the bond type you want from the toolbar, then clicking on the atoms you want to connect. To change the length or angle of the bond, click and drag on the bond.

5. Add charges to the atoms by selecting the "Charge" tool from the toolbar, then clicking on the atom you want to add a charge to. You can also type the charge directly on the canvas next to the atom.

6. Add lone pairs to the atoms by selecting the "Lone Pair" tool from the toolbar, then clicking on the atom you want to add a lone pair to.

7. Add functional groups to the chemical structure by selecting them from the toolbar, then clicking on the appropriate atom or bond.

8. Save your chemical structure by selecting "File" > "Save As" and choosing the file format you want.

Application:

- *Drawing chemical structures*: ChemDraw provides an intuitive and user-friendly interface for drawing and creating chemical structures and reactions.

- *Designing molecules*: ChemDraw is widely used for designing molecules, creating new compounds and exploring their properties.
- *Creating professional-looking scientific illustrations*: ChemDraw provides a range of tools for creating high-quality chemical diagrams and illustrations that can be used for research papers, presentations, and publications.
- *Creating 3D molecular models*: ChemDraw can be used to create 3D models of molecules, which can help researchers to better understand the structure and properties of molecules.
- *Calculating properties*: ChemDraw offers various tools for calculating properties such as molecular weight, mass spectra, and UV/Vis spectra.
- *Sharing data with other applications*: ChemDraw can export chemical structures in various formats, which can be used in other applications such as Microsoft Office, Adobe Illustrator, and other scientific software.
- *Teaching and learning*: ChemDraw is used by educators and students to create and share chemical structures, reactions, and other illustrations in chemistry and related fields. It is widely used in chemistry courses at all levels, from high school to graduate school.

Result:

The chemical structures of given aliphatic and aromatic organic compounds are drawn using Chem Draw software.

Observation

Draw the following chemical structures using Chem Draw software:

1) Diclofenac, 2) Ascorbic Acid, 3) Glyphosate and 4) 1-Hydroxypyrene

Experiment A1 (ECE) : Synthesis of Polyurethane

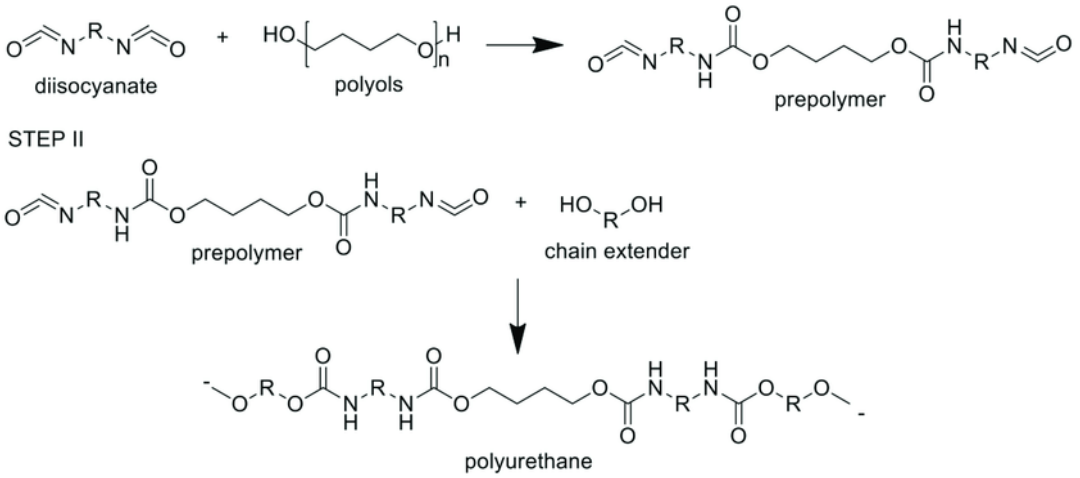
Significance of experiment

Polyurethanes are organic polymers that are formed by reacting a polyol (an alcohol with more than two reactive hydroxyl groups per molecule) with a diisocyanate or a polymeric isocyanate in the presence of suitable catalysts and additives. Polyurethanes are a versatile class of polymers with great control over their physicochemical properties based on the chemical composition. Flexible polyurethane forms are used in upholstery, mattresses, chemical-resistant coatings, adhesives, sealants, and packaging. Rigid foams are used in insulation for buildings, water heaters, refrigerated transport, and commercial and residential refrigerators. In medical practice, polyurethane is used in implants. Complications of polyurethane implants are not uncommon, mainly due to foreign-body reactions, such as with microporous polyurethane (Mitrathane) cardiac patch implants. Segmented PURs are designed with well-defined degradation and mechanical properties combined with excellent biocompatibility that makes them attractive for the development of drug delivery, tissue engineering, and medical devices. Various PURs including PEURs, poly(ester urethanes), PCURs, PSURs, surface-modified PURs, and composite PURs have been developed for a variety of biomedical applications. Many research efforts are continued in the development of PURs for specific drug delivery and tissue regeneration application with a particular emphasis on biocompatibility and biodegradability.

Aim: To synthesize polyurethane from polyol and diisocyanate.

Theory: Diisocyanates are a family of chemicals used to make a wide range of polyurethane products. Polyurethane foam have wide range of applications in everyday products, including furniture, bedding, carpet underlay, and packaging.

Polyols, by themselves, can also be considered polymers. However, for the production of polyurethanes, the chain length of the polyol and the functionality of the polyol must be controlled. These factors are known to contribute to the properties of the final polyurethane products. For example, the polyols whose molecular weights lie in the hundreds yield rigid polyurethanes whereas the polyols whose molecular weights lie in the thousands yield relatively flexible polyurethanes. Thus, the chain length of the polyol used can be considered a factor that contributes to the flexibility of the polyurethane product.



Materials: Toluene diisocyanate (TDI) or methylene diphenyl diisocyanate (MDI), Ethylene glycol or Polytetramethylene Ether Glycol, 1,4-butanediol.

Procedures: Take 5 ml of polyol in a disposable container. Mix it with 5 ml of diisocyanate and make sure the substances mixes well using stirrer or glass rod. Slowly add 5 ml of chain extender to the solution and mix it properly. Transfer reaction mixture immediately to a mold and remove the air baubles using a hot air gun.

Applications:

1. The primary application of polyurethane is in the production of foams. These foams are used in a variety of materials such as upholstery fabrics, domestic furniture, and refrigerator sheets.
2. Polyurethane mouldings are also used in columns and door frames.
3. The low-density foams of polyurethane which exhibit flexibility are widely used in mattresses and other forms of bedding. They are also used in automobile seats and upholstery.
4. The low-density elastomers of polyurethane are widely used in the footwear industry.

Result: Weight of polyurethane obtained = ……g

Experiment A3 (CSE & ECE): Synthesis of Iron Oxide nanoparticles

Significance of experiment

Nanoparticles (NPs) are at the forefront of rapid development in nanotechnology. Their exclusive size-dependent properties make these materials indispensable and superior in many areas of human activities.1 Being the most current transition metal in the Earth's crust, iron stands as the backbone of current infrastructure. Generally, iron oxides are prevalent, widely used as they are inexpensive, and play an imperative role in many biological and geological processes. They are also extensively used by humans, eg, as iron ores in thermite, catalysts, durable pigments (coatings, paints, and colored concretes), and hemoglobin. The most common forms of iron oxides in nature are magnetite (Fe_3O_4), hematite (α-Fe_2O_3) and *maghemite (γ-Fe_2O_3). These oxides are also very important in the field of scientific technology. In this experiment we are going learn about how to synthesis of γ-Fe_2O_3 nanoparticles. This experiment will also make the students familiarize with the synthesis of nanomaterials through wet chemistry methods.*

Aim: Synthesis of iron oxide nanoparticles vis co-precipitation method under alkaline medium.

Theory: Numerous chemical, physical and biological routes have been utilized in order to produce appropriate surface chemistry of magnetic nanoparticles. The common chemical method including ***coprecipitation***, sol-gel, hydrothermal, microemulsion and thermal decomposition. Among these methods, coprecipitation method is the most promising and cost effective method to produce γ-Fe_2O_3 nanoparticles due to its simplicity and high productivity. The characteristics of final product such as surface response, particle size and shape of the IONPs depends upon preparation technique. Only the nano-sized **maghemite (γ-Fe_2O_3)** nanoparticles with appropriate size, shape and purity has noteworthy impact in diverse biomedical applications and different scientific research areas.

Materials: Iron (II) chloride ($FeCl_2.4H_2O$, 99% pure), Iron (III) chloride ($FeCl_3$, 98% pure, anhydrous), and NH_4OH.

Procedures: In this experiment, γ-Fe_2O_3 nanoparticles have been prepared by co-precipitation method. 0.7954 g of iron (II) chloride /sulphide, and 1.2974 g of iron (III) chloride, $FeCl_3$ were each dissolved in distilled water. The solution was then stirred at 50°C for 30 minutes. After that, ammonia is added to the solution and the mixed solution was

washed/separated by filtering or centrifugation. The separated precipitate was dried in the oven at temperature of 100°C for 24 hours. Finally, the dried dark brown precipitated sample was collected and crushed into powder form by using a pestle and mortar. The chemical balanced equation for the reaction $FeCl_2.4H_2O$ and $FeCl_3$ is shown in Equation 1-3 where γ-Fe_2O_3 was the final product and water was the by-product.

$$\mathbf{Fe^{2+} + 2Fe^{3+} + 8\ OH^- \rightarrow Fe_3O_4 + 4H_2O} \quad (1)$$

$$\mathbf{Fe_3O_4 + 0.25\ O_2 + 4.5\ H_2O \rightarrow 3\ Fe(OH)_3} \quad (2)$$

$$\mathbf{2\ Fe(OH)_3 \rightarrow \gamma\text{-}Fe_2O_3 + 3H_2O} \quad (3)$$

Characterization: γ-Fe_2O_3 nanoparticles formed from all the processing conditions can be systematically characterized in terms of functional groups, phase composition and crystallite size as well as morphology structure by using Fourier Transform Infrared Spectroscopy (FTIR), X-Ray Diffraction (XRD) and Scanning Electron Microscopy (SEM) respectively.

Applications: Development super paramagnetic materials and Super capacitor materials. Also used for catalysts, durable pigments, coatings, paints and colored concretes.

Result: Weight of iron oxide nanoparticle obtained = ……g

Experiment B1 (CSE & ECE): Conductometric estimation of acid mixture

Significance of the experiment:

Conductivity (or specific conductance) of an electrolyte solution is a measure of its ability to conduct electricity.1 The SI unit of conductivity is siemens per meter (S/m). Conductivity measurements are used routinely in many industrial and environmental applications as a fast, inexpensive and reliable way of measuring the ionic content in a solution. For example, the measurement of product conductivity is a typical way to monitor and continuously trend the performance of water purification systems. In many cases, conductivity is linked directly to the total dissolved solids (T.D.S.). High quality deionized water has a conductivity of about 5.5 µS/m, typical drinking water in the range of 5-50 mS/m, while sea water about 5 S/m. (i.e., sea water's conductivity is one million times higher than that of de-ionized water).Conductivity is traditionally determined by measuring the AC resistance of the solution between two electrodes. Dilute solutions follow Kohlrausch's Laws of concentration dependence and ionic contributions. Lars Onsager gave a theoretical explanation of Kohlrausch's law by extending Debye–Hückel theory

Aim: To estimate **HCl** and **CH_3COOH** by conductometry using standard sodium hydroxide solution.

Principle: Conductometry is based on Ohm's law.

- Ohm's law states that the current I (amperes) flowing in a conductor is directly proportional to the applied electromotive force, E (volts), and inversely proportional to the resistance R (ohms) of the conductor.

$$I = \frac{E}{R}$$

- The reciprocal of the resistance is called the conductance. The resistance of a homogeneous material of uniform cross-section with an area of a sq. cm. and length *l* cm is given by

$$R = \frac{\rho \times l}{a}; \quad \mathbf{k = C\,[l/a]}$$

Where 'ρ' is the specific resistance. The reciprocal of the specific resistance is termed the specific conductance, κ.

Specific conductance: Specific conductance of a solution is defined as the conductance of a solution present between two parallel electrodes which have 1 cm^2 area of cross section and which have kept 1 cm apart.

During the conductometric titration process, one ion is replaced with another and the difference in the ionic conductivities of these ions directly impacts the overall electrolytic conductivity of the solution. Therefore, the conductance of the solution depends on the number of mobility of ions. The equivalence point is determined graphically by plotting conductance against titer values.

Let V_1 and V_2 ml be the volume of NaOH corresponding to the first and second neutralization respectively then 'a' ml of NaOH = HCl and (V_2 - V_1) ml of NaOH = CH_3COOH.

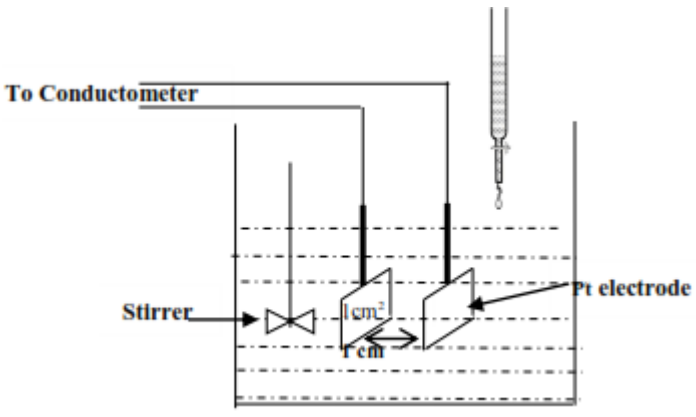
$$H^+ + Cl^- + Na^+ + OH^- \rightarrow Na^+ + Cl^- + H_2O$$
analyte titrant

High ionic conductivity: H^+ and OH^-

Low ionic conductivity: Na^+ and Cl^-

$$CH_3COOH + Na^+ + OH^- \rightarrow CH_3COO^- + Na^+ + H_2O$$
analyte titrant

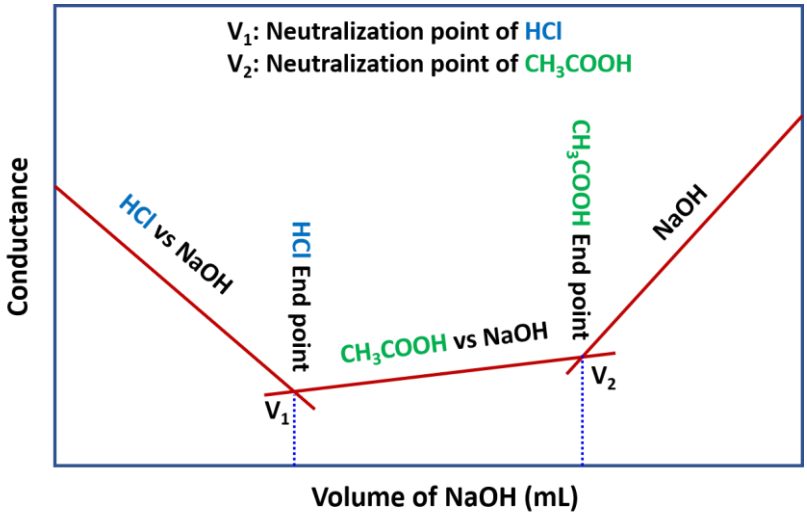
Instrumentation: Conductometer consists of a conductivity cell having two platinum electrodes and a conductance measuring device. The two electrodes have a unit area of cross section and are placed unit distance apart. A simple arrangement of conductometric titration is depicted in the figure. The solution to be titrated is taken in the beaker.



Procedure: Exactly 50 ml of the given acid mixture was pipette out into a clean 100ml beaker, the conductivity cell was immersed into the beaker containing acid mixture. The conductivity was noted before adding the titrant, then 1N NaOH solution was added from the burette in the increment of 1 mL and the conductivity was noted at each time. Then the graph was plotted with the volume of NaOH along X-axis and the conductivity along the Y-axis. From the graph, the volume of NaOH required to neutralize the acid mixture was found out. Using the normality equation, $\mathbf{N_1V_1 = N_2V_2}$, we can find out concentrations of strong acid and weak acid.

Applications:

i) Mixture of acids with a Strong base [(HCl+CH_3COOH) vs NaOH]: Conductometric titration may be applied for the determination of acids present in a mixture.[1,2] In the titration of a mixture of week acid (CH_3COOH) and strong acid (HCl) with a strong base (NaOH), the conductance decreases upon adding NaOH to acid mixture owing to the substitution of highly mobile H^+ ions by the less mobile Na^+ ions. This trend continues till all the H^+ ions of HCl are replaced i.e., the strong acid is neutralized. Continued addition of NaOH raises the conductance moderately, as the weak acid (CH_3COOH) is converted into its salt (CH_3COONa). Further addition of NaOH raises the conductance steeply due to the presence of OH^- ions. The titration curve in the graph given determines the location of the equivalence points.



Advantages:

i) Mixture of acids can be titrated more accurately by conductometric titration.

ii) Conductometric titrations may be applied where potentiometric methods fail.

iii) Accurate in dilute solution as well as in more concentrated solution.

iv) It can be employed with colored solutions.

v) Very weak acids which cannot be titrated potentiometrically in aqueous solutions can be titrated conductometrically with relative ease.

Result:

Weight of HCl in given acid mixture =……..g /L

Weight of CH_3COOH in given acid mixture =……..g/L

Experiment: Observation and calculations

Sl No.	Volume of NaOH (mL)	Conductivity ($Ohm^{-1}cm^{-1}$)
1		
2		
3		
4		
5		
...		
...		
....		
....		

Normality of NaOH solution =..... (Will be provided to you)

Normality of HCl = $\frac{Normallity\ of\ NaOH\ x\ Volume\ of NaOH\ (V1)}{Volume\ of HCl\ (50)}$ = ……..N (c)

Weight of $\frac{HCl}{Liter\ (L)}$ = 'c' x Equivalent weight of HCl (36.5)

= ……..g

Normality of CH_3COOH = $\frac{Normality\ of NaOH\ x\ Volume\ of NaOH\ (V2-V1)}{Volume of CH3COOH\ (50)}$ =……N (d)

Weight of $\frac{CH3COOH}{Liter\ (L)}$ = 'd' x Equivalent weight of CH_3COOH (60.5)

= ……..g of CH_3COOH

Result:

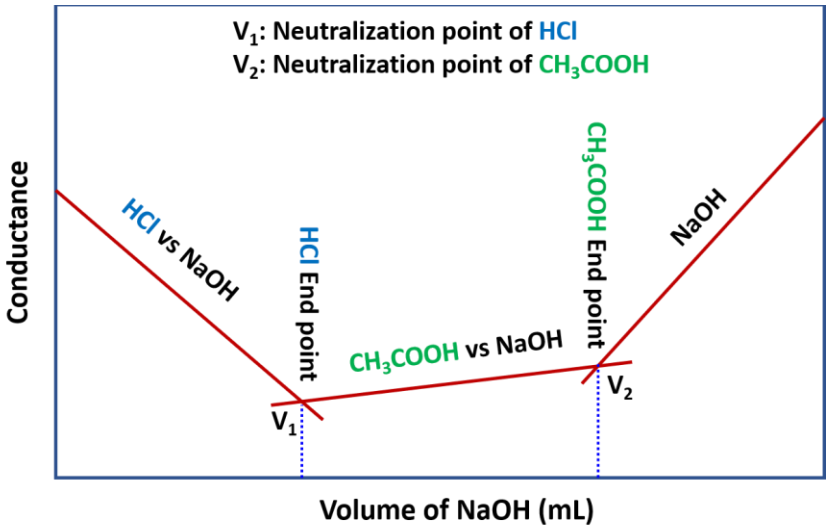
Weight of HCl in given acid mixture =……..g /L

Weight of CH_3COOH in given acid mixture =……..g/L

Model procedure

Experiment B1: Conductometric estimation of acid mixture

Pipette out 50 mL acid mixture into a beaker. Immerse the conductivity cell into it. Connect the conductivity cell to a conductivity meter and measure the conductance by adding NaOH from the burette by an increment of 1 mL. Plot a graph of conductance against volume of NaOH. Determine the two neutralization points from the graph as shown below and find the weight of HCl and CH_3COOH.



Experiment B2 (CSE & ECE): Potentiometric estimation of FAS using $K_2Cr_2O_7$

Significance of experiment:

In potentiometry we measure the potential of an electrochemical cell under static conditions. Because no current or only a negligible current flows through the electrochemical cell, its composition remains unchanged. For this reason, potentiometry is a useful quantitative method. The first quantitative potentiometric applications appeared soon after the formulation, in 1889, of the Nernst equation, which relates an electrochemical cell's potential to the concentration of electroactive species in the cell. Turbid, fluorescent, opaque or coloured solutions can be titrated. Mixture of solutions or very dilute solutions can be titrated. The results are more accurate because the actual end point is determined graphically.

Aim: To estimate FAS potentiometrically using standard potassium dichromate solution.

Theory: The estimation of concentration of substances in solution by the measurement of emf is known as potentiometric titration. Here, emphasis is laid on the changes in emf of an electrolytic cell as a titrant of known concentration is added.

When a metal M is immersed in a solution containing its own ions M^{n+}, the electrode potential is given by Nernst equation.

$$E_{cell} = E^{\circ} + \frac{0.0591}{n} \log [M^{+n}]$$

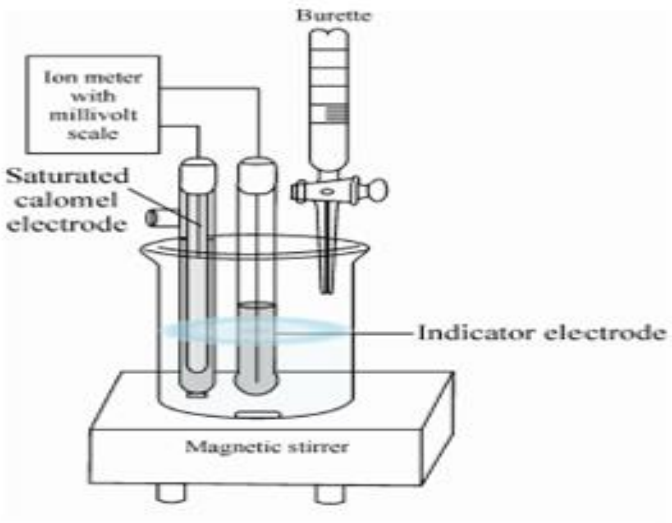
Thus, the concentration can be calculated, provided E^{o} of the electrode is known. If an electrode of the metal reversible with respect to the corresponding ions is placed in the solution, the potential will vary throughout the titration. Initially, the change in potential will be small. At the equivalence point, there will be a steep rise in the potential. The equivalence point can be determined by plotting the change in potential against volume of titrant added.

Instrumentation: A potentiometer consists of: (i) Calomel electrode as a reference electrode, (ii) Platinum electrode as an indicator electrode, (iii) a device for measuring the potential and (iv) magnetic stirrer.

Procedure: Pipette out 25ml of the given FAS solution into a clean beaker and add 1 test tube of dil H_2SO_4. Immerse a platinum electrode and saturated calomel electrode and connect it to

a potentiometer. Fill a clean burette with the given standard $K_2Cr_2O_7$ solution. Add $K_2Cr_2O_7$ from the burette in the increment of 0.5ml of and mix the solution. Measure the emf of the solution after every addition till there is a rise in the emf. Take few more readings. Plot a graph of $\Delta E/\Delta V$ (ordinate) against volume of $K_2Cr_2O_7$. From this graph volume of $K_2Cr_2O_7$ at equivalent point is found out. Following reaction takes place during titration.

$$Cr_2O_7^{2-} + 14\ H^+ + Fe^{2+} \rightarrow Fe^{3+} + Cr^{3+} + 7H_2O$$



Advantages of potentiometric titrations are:

(i) They give results more reliable than those obtained from titrations using indicators
(ii) The method is applicable to both colored and turbid solutions also

Result: (i) Normality of FAS (N_{FAS}) = ……

(ii) Weight of FAS present in given solution = …… g

Experiment: Observation and calculations

Volume of $K_2Cr_2O_7$ added (mL)	E (mV)	ΔE	ΔV	$\frac{\Delta E}{\Delta V}$
		---	--	--

From the graph, volume of $K_2Cr_2O_7$ at the equivalence point = …… (x) mL

Normality of $K_2Cr_2O_7$ = …… (a) (will be provided to you)

Normality of FAS (N_{FAS})= $\frac{Normality\ X\ Volume\ of K2Cr2O7}{Volume\ of FAS}$ = $\frac{a\ x\ X}{25}$ = ……(say b)

Weight of FAS / L = N_{FAS} x equivalent weight of FAS = b x 392 = …….g (say c)

Weight of FAS / 25 mL = $\frac{c}{40}$ =…….g

Result: (i) Normality of FAS (N_{FAS}) = ……

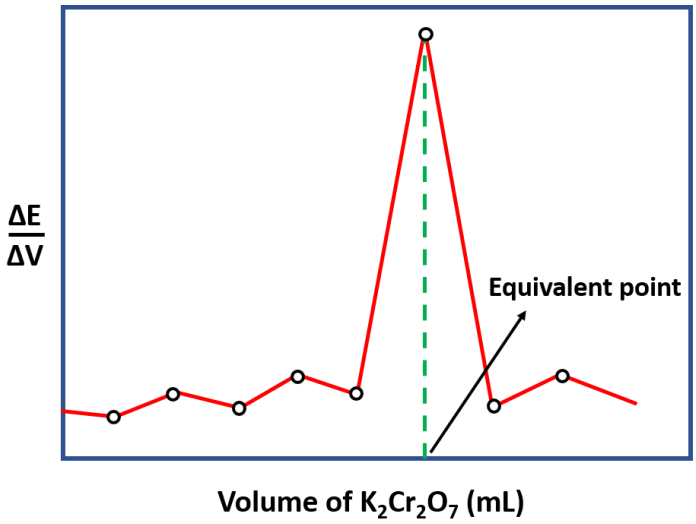
(ii) Weight of FAS present in given solution = …… g

Model procedure

Experiment B2: Potentiometric estimation of FAS using $K_2Cr_2O_7$

Pipette out 25ml of FAS into a beaker. Add 1 test tube dil. H_2SO_4, immerse calomel electrode & platinum electrode into it. Connect the assembly to a potentiometer and measure the potential by adding $K_2Cr_2O_7$ in the increments of 0.5 ml.

Plot graph $\Delta E/\Delta V$ against volume of $K_2Cr_2O_7$, and determine the equivalence point. From the normality and volume of $K_2Cr_2O_7$ solution, calculate the normality and the weight of FAS in the given solution.



Experiment B3 (CSE & ECE): Determination of pKa of vinegar using pH sensor (Glass electrode)

Significance of the experiment:

The acid dissociation constant (pKa) is among the most frequently used physicochemical parameters, and its determination is of interest to a wide range of research fields.1,2 The related concept of the acid dissociation constant (pKa) as a substance property is recognized as being among the most commonly used parameters in modern-day chemistry. Both pH and pKa are essential for understanding the behavior of chemical substances in everyday life. The quantitative behaviour of acids and bases in solution can be understood only if their pKa values are known.3 In particular, the pH of a solution can be predicted when the analytical concentration and pKa values of all acids and bases are known; conversely, it is possible to calculate the equilibrium concentration of the acids and bases in solution when the pH is known. These calculations find application in many different areas of chemistry, biology, medicine, and geology. For example, many compounds used for medication are weak acids or bases, and a knowledge of the pKa values, together with the water–octanol partition coefficient, can be used for estimating the extent to which the compound enters the blood stream. Acid dissociation constants are also essential in aquatic chemistry and chemical oceanography, where the acidity of water plays a fundamental role. In living organisms, acid-base homeostasis and enzyme kinetics are dependent on the pKa values of the many acids and bases present in the cell and in the body. In chemistry, a knowledge of pKa values is necessary for the preparation of buffer solutions and is also a prerequisite for a quantitative understanding of the interaction between acids or bases and metal ions to form complexes. Experimentally, pKa values can be determined by potentiometric (pH) titration using pH meter.pH meter is one of the most innovative equipments of 20th century. This instrument is an essential component of any pharmaceutical lab, clinical lab, research labs of biochemistry, molecular biology and chemistry. The main application of pH meter is determination of pH of buffers. Due to its accuracy, this is an essential component in every place where aqueous buffers are used.

Aim: To determine pK_a value of vinegar (a weak acid) using pH meter.

Principle: The strength of an acid is experimentally measured by determining its equilibrium constant or dissociation constant (K_a). Since strong acids are strong electrolytes, they are ionized almost completely in aqueous solutions. It is not meaningful to study the ionic equilibrium of strong acids and calculate their equilibrium constants, as the un-ionised form is present to such a small extent. Hence, the study of ionic equilibrium and calculation of K_a is applicable only to weak acids.

Vinegar (Acetic acid) dissociate into ions but not completely as shown below.

$$CH_3COOH\ (aq.) + H_2O\ (l) \rightleftharpoons H_3O^+\ (aq.) + CH_3COOH\ (aq.)$$

$$\boldsymbol{Ka = \frac{[H3O^+][CH3COO^-]}{[CH3COOH]}}$$

pK_a is a modern method of expressing acid strength $\mathbf{pK_a = -\log [K_a]}$

pK_a is determined by measuring the changes in pH of acid solution at different amounts of the base added. During the titration of an acid with a base, the pH of the solution rises gradually at first, then more rapidly and until at the equivalence point, there is a very sharp increase in pH for a very small quantity of added base. Once past the equivalence point, the pH increases only slightly on addition of excess base. The titration curve is obtained by plotting changes in pH at different amounts of the base added and the equivalence point is determined.

According to Henderson- Hasselbalch equation,

$$\mathbf{pH = pK_a + \log \frac{[salt]}{[acid]}}$$

At half equivalence point, [salt] = [acid] and therefore, pH at half equivalence point gives the $\mathbf{p}K_a$ of weak acid.

Procedure:

Pipette out of 25 cm^3 of the given vinegar solution into a beaker. Immerse a glass electrode-calomel electrode assembly into the acid and connect the cell to a pH meter. Measure the pH of the. Fill a burette with the sodium hydroxide solution, add 0.5 ml increments of NaOH and Stir the solution thoroughly and measure the pH after each addition, continue till there is rapid increase in pH and take another 4 or 5 values after rapid increase.

Plot a graph of pH (ordinate) against the volume of sodium hydroxide added (absicissa). Determine the equivalence point and hence the pH at half equivalence point. This gives the $\mathbf{p}K_a$ value of the acid.

Result:

The pK_a value of the given vinegar is = ...

Experiment: Observation and Calculations

Volume of NaOH (mL)	pH	ΔpH	ΔV	ΔpH/ΔV
		--	--	--

Equivalence point = ……

Half-equivalence point = ……

pH at half-equivalence point = ……

pK_a of weak acid = pH at half-equivalence point

Result:

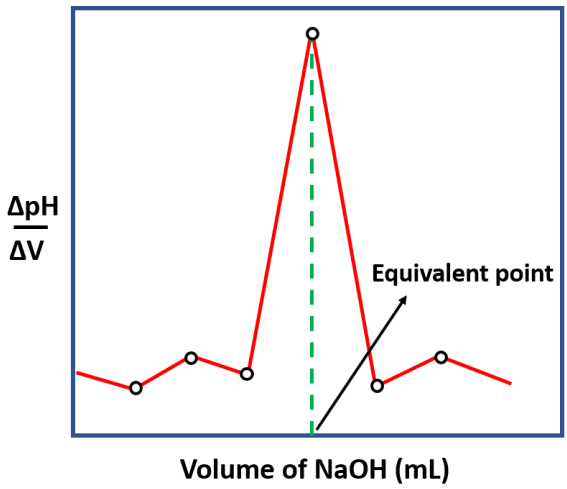
The pK_a value of the given vinegar is = ...

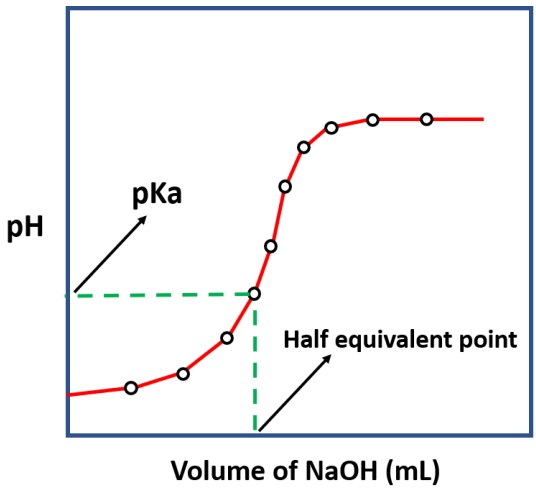
Model procedure

Experiment B3 (CSE & ECE): Determination of pKa of vinegar using pH sensor (Glass electrode)

Pipette out 25 ml of given vinegar solution into a beaker. Immerse the glass electrode + Calomel electrode assembly into it. Connect the electrodes to pH meter and measure the pH. Now add NaOH from the burette in increments of 0.5 ml and measure the pH after each addition.

Plot graph ΔpH/ ΔV against the volume of NaOH. Determine the equivalence point. Plot another graph of pH against volume of NaOH and determine pKa of the given weak acid as shown below.





Experiment B5 (CSE & ECE): Estimation of total hardness of water by EDTA method

Significance of experiment

Water from many natural sources contain a variety of dissolved salts which comprise ions such as Na+, K+, Ca2+, Mg2+, Cl-, HCO3-.1According to U.S. geological survey any water that has less than 60 mg/L of CaCO3 mass equivalents (i.e. less concentrations of Ca2+ and Mg2+ ions) is considered soft water.2Higher concentrations of Ca2+ and Mg2+ ions in water render hardness and such water is referred to as hard water. Hard water may lead to (a) inefficient and dysfunctional boilers due to formation of scales; (b) excessive consumption of soap that arises due to the reaction of sodium soap with multivalent metal cations in turn leading tothe formation of precipitate and the loss of its surfactant property. To increase the energy efficiency in industries3 and to minimize wastage of soap for cleaning purposes it is essential to use soft water, which therefore means there is a need for a method to identify which is hard and which is soft water? And to estimate how hard the tested water is? This experiment is all about one such easy methods.

Aim: To determine the total hardness of given water sample using standard Na_2EDTA solution

Principle: Hardness of water is due to the presence of calcium and magnesium salts in water. Ethylene diaminetetraaceticacid (EDTA) forms complexes with a large number of cations including Ca^{2+} and Mg^{2+} ions. Accordingly, it is possible to determine the total hardness of water using EDTA reagent. The EDTA molecule (H4Y) has two easily replaceable hydrogen atoms and resulting ion after ionization may be represented as H2Y2-. It forms complexes with metal ions as follows.

$$M^{2+} + H_2Y^{2-} \longrightarrow MY^{2-} + 2H^{+} \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots (1)$$

Where M^{2+} is Ca^{2+} and Mg^{2+} present in water. Reaction (1) can be carried out quantitatively at a pH of 10 using Eriochrome Black T indicator. Since the reaction involves the liberation of H^{+} ions, a buffer mixture has to be used to maintain pH of 10. The buffer mixture used in the titration is NH_3-NH_4Cl. The hardness of water is usually expressed in terms of ppm (parts per million) of $CaCO_3$. Since EDTA (free acid) is sparingly soluble, its disodium salt, Na_2H_2Y is used for preparing the reagent.

Procedure:

Part A: Preparation of standard solution of disodium salt of Na_2EDTA

Weigh Na_2EDTA salt accurately and transfer to a 250 mL volumetric flask through a funnel. Add around 5 mL of ammonia (NH_3) solution. Dissolve the crystals in ion exchange water, dilute up to the mark and mix well. Calculate the molarity of Na_2EDTA solution.

Part B: Determination of total hardness of given water sample

Pipette out 25 mL of the given water sample into a clean conical flask. Add 3 mL of NH_3-NH_4Cl buffer and a pinch of EBT indicator. Titrate against Na2EDTA solution till the color changes sharply from wine red to clear blue. Perform the titration slowly towards the end point and repeat the experiment for agreeing values.Steel nail of known weight dipped in tap water

Result: The total hardness of given water sample is = ----------ppm of $CaCO_3$

Experiment B4: Observation and Calculations

Part A: Preparation of standard solution of disodium salt of EDTA (Na_2EDTA).

1. Weight of bottle + Na_2EDTA salt = W_2= ---------- g

2. Weight of empty bottle = W_1 = ----------g

3. Weight of Na_2EDTA salt = W_2-W_1= ---------- g

$$Molarity\ of Na2EDTA\ = \frac{Weight\ of\ the\ salt\ (W2 - W1)\ X\ 4}{Molar\ mass of Na2EDTA\ (372.24g)}$$ ---- (`Z')

Part B: Determination of total hardness of given water sample

Burette Reading	**Trial I**	**Trial II**	**Trial III**
Final Reading			
Initial Reading			
Volume of Na_2EDTA run down (mL)			

Volume of Na_2EDTA consumed = ---------- mL (a)

1000 mL of 1M Na_2EDTA = 100 g of $CaCO_3$ (molar mass of $CaCO_3$ = 100 g)

`a' mL of `Z'M Na_2EDTA = $\frac{100\ X\ Z\ X\ a}{1000\ X\ 1}$ = ---------- g of $CaCO_3$ (b)

Amount of $CaCO_3$ in 25 mL of hard water solution = ---------- g (b)

Total hardness of the given water sample ---- ppm of $CaCO_3$

= $\frac{b\ X\ 10{,}00{,}000}{25}$ =

Result: The total hardness of given water sample is = ----------ppm of $CaCO_3$

Model procedure

Experiment B5 (CSE & ECE): Estimation of total hardness of water by EDTA method

♦ Part A

Weigh out the given Na_2EDTA crystals accurately into a 250 ml volumetric flask and add 5 ml of Ammonia solution. Dissolve in distilled water and make up to the mark. Mix well and find the Molarity.

$$\text{Molarity of } Na_2EDTA = \frac{\text{Weight of } Na_2EDTA \times 4}{\text{Molecular weight of EDTA (372.24)}}.$$

♦ Part B

Burette : Standard Na_2EDTA solution

Conical flask: 25ml of water sample + 3ml of (NH_3NH_4Cl) buffer solution to maintain pH =10

Indicator : Eriochrome Black-T (a pinch)

End point : Wine red to clear blue

Conclusion : From the volume of Na_2EDTA consumed calculate the total hardness of given water sample

Experiment C1(CSE & ECE): Estimation of Copper present in electroplating effluent by optical sensor (colorimetry)

Significance of experiment:

Electroplating industrial activities are among the major pollutants to the environment. Such activities led to the formation of hazardous wastewater associated with toxic heavy metals that threaten the environment and aquatic lives. The electroplating effluents are monitored using various analytical techniques for the presence of these toxic metals. Colorimetry is an important analytical technique used to monitor electroplating effluents. It is used in chemistry and in other sorts of places such as in industries like colour printing, textile manufacturing, paint manufacturing and in food industries. Colorimetry is also used in asprin. Colorimetry can detect the smallest colour difference that the human eye cannot pick up. Under the action of chemical agents, samples develop a specific colour that shows the concentration of the substance being tested. Colorimetry is just one of the types of photometric analysis techniques i.e. it is a way of measuring light. Colorimetry can be used to find out the concentration of any coloured substance. Such as in Food & Beverage Quality Control - Alpha Amylase Activity - Milk Quality - Miscellaneous Quality Tests etc. Colorimetry measurements are made by using a light which passes through a colour filter. The light then passes through a little box (cuvette) with the actual chemical substance. The light leaving the actual sample should be less than the light that actually entered the compound. The loss of light always reflects the concentration of the compound.

Aim: To determine copper present in electroplating effluent by optical sensor (colorimeter)

Principle: When a monochromatic light of intensity I_0 is incident on a transparent medium, a part I_a of is absorbed, a part I_r is reflected and the remaining part is transmitted I_t.

$$\mathbf{I_0 = I_a + I_r + I_t}$$

For a glass-air interface I_r is negligible, therefore,

$$\mathbf{I_0 = I_a + I_t}$$

$\mathbf{I_t / I_0 = T}$ called the transmittance, $\mathbf{\log 1/T = \log I_0 / I_t}$ is called the absorbance or optical density.

Colorimetry measurements are based on ***Beer-Lambert's law***. This law gives the relation between absorbance A, concentration c (expressed in mo1/dm^3) and path length t, (expressed in cm).

Beer-Lambert's law: When monochromatic light passes through a transparent medium, the amount of light absorbed is directly proportional to the concentration and path length of the solution.

A C

$$A = \log I_0/I_t = \varepsilon ct$$

Where **ε** is the molar extinction coefficient, **c** is the concentration, **t** is the path length and is a constant for a given substance at a given wavelength. If the length is kept constant (t),

A ∝ c

Hence a plot of absorbance against concentration gives a straight line.

Instrumentation: The instrument used to measure the absorbance of a solution is called photoelectric colorimeter.

It consists of

(i) Tungsten lamp as the light source.

(ii) A filter which provides the desired wavelength range wherein the solution gives the maximum absorbance.

(iii) A sample cell

(iv) A photocell detector

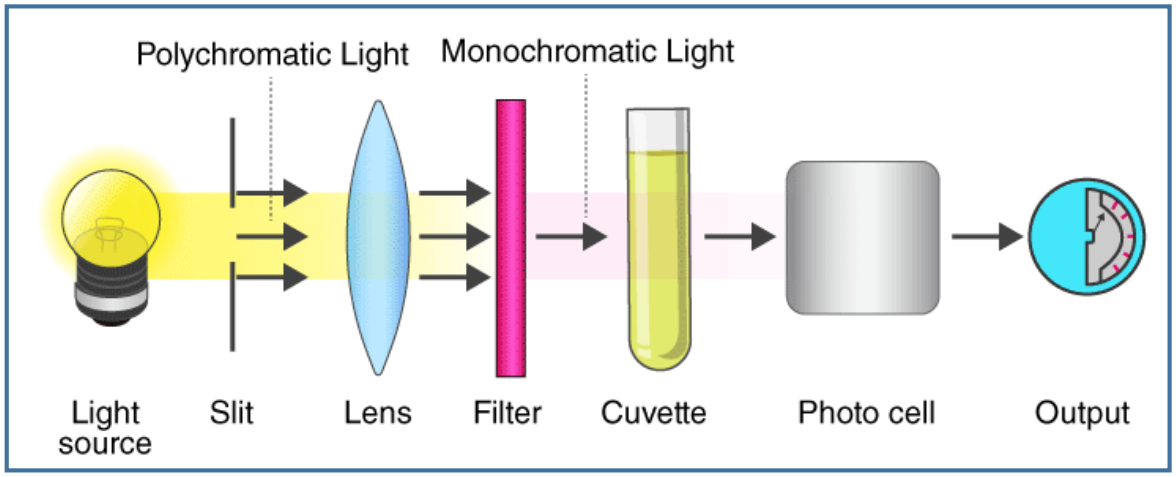


Fig: Schematic layout of colorimeter

Application: Colorimetry is a versatile method of determining the concentration of metals and nonmetals in small quantities in ores, soil, samples and alloys. eg., Colorimetric estimation of Cu in $CuSO_4$.

Procedure: Transfer the given copper sulphate solution (stock solution) to a burette and draw out **2**, **4**, **6**, **8** and **10** mL of the solution into a 50 mL volumetric flask. Add 5 mL of ammonia solution to each of them and dilute up to the mark with ion exchange water. Stopper the flasks and mix the solutions well. add 5 mL of ammonia solution to the given test solution and then dilute up to the mark with ion exchange water and mix well.

Prepare a blank solution by diluting 5 mL of ammonia solution in a 50 mL measuring flask up to the mark with ion exchange water and mixing well. Measure the absorbance of the solutions against the blank at 620 nm using a photoelectric colorimeter. Tabulate the readings as shown. Draw a calibration curve by plotting the absorbance against the volume of copper sulphate solution. Using the calibration curve, find out the volume of copper sulphate solution given i.e., the volume of the test solution and calculate the amount of copper in the given solution.

Advantage

i) Colorimetry gives accurate results at low concentrations.

ii) Colorimetry is also applied to biological samples.

Result:

From graph, volume of copper sulfate in test solution = ……..mL

The weight of copper in the given test solution = …….. mg

Experiment: Observation and calculations

Volume of $CuSO_4$ (mL)	Absorbance (Optical density)
2	
4	
6	
8	
10	
Test solution	

1000 mL of stock solution contains `**a**' g of $CuSO_4$ (`**a**' value will be given).

249.54 g of $CuSO_4$ = 63.54 g of Cu.

`**a**' g of $CuSO_4$ $= \frac{63.54}{249.54} \times \mathbf{a}$ = ... g of Cu in 1000 mL.

1 mL of $CuSO_4$ =..... mg of Cu (say **b**)

`**c**' mL of test solution = b x c = ...mg of Cu

Result:

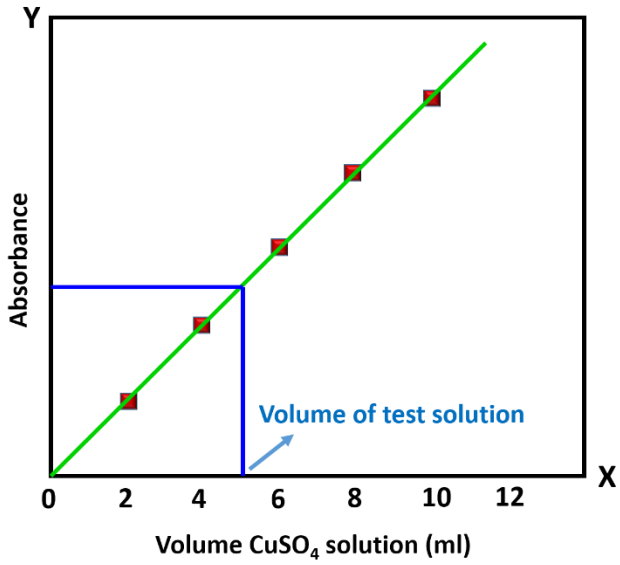
From graph, volume of copper sulfate in test solution = ...mL

The weight of copper in the given test solution = ... mg

Model procedure

Experiment C1: Estimation of Copper present in electroplating effluent by optical sensor (colorimetry)

Draw out 2, 4, 6, 8, and 10 ml of the copper sulphate solution into a 50 ml volumetric flask. Add 5 ml of ammonia solution to each of them, dilute up to the mark with distilled water, and mix well. Prepare a blank solution by diluting 5 ml of ammonia solution in 50 ml volumetric flasks. For the test solution add 5 ml of NH_3 and make up to the mark. Measure the absorbance of each of these against a blank solution at 620 nm. Plot a graph of absorbance (OD) against the volume of copper sulphate solution, determine the volume of copper sulphate solution in the test sample as shown in the figure, and find the amount of copper present in it.



Experiment C2 (CSE & ECE): Determination of Viscosity coefficient of lubricant (Ostwald's viscometer)

Significance of the experiment:

Viscosity gives us precisely how fast a liquid/fluid flows. Ever wondered why chocolate melts if it is not kept at cold temperature. The reason for this is the change in viscosity of the chocolate syrup with temperature. Viscosity is sensitive to temperature and in some fluids its value doubles for a mere 5°C increase in temperature. A measure of viscosity is very important in the food industry to increase the consistency, quality, production efficiency, and texture in foods. Similarly, viscosity is precisely maintained at specific value in paints and varnishes so that they can be evenly and smoothly applied over the surface with brush roller or a paintbrush. For easy flow of the products such as toothpaste, syrups, and lubrication oils they should have a particular value of viscosity to be useful. Viscosity coefficient is same as dynamic viscosity. The ratio of shearing stress to the velocity gradient gives viscosity coefficient. The SI unit of dynamic viscosity is pascal second and cgs unit is poise. 1 millipoise = 1/1000 poise and 1 poise = 1/10 pascal second (Poise unit is named after Jean Poiseuille, scientist who first observed the factors that influence the flow of fluids in circular tubes and derived an equation for Viscosity). In this particular experiment you are only interested in the flow of non-turbulent (steady) and non-pulsatile (no periodic variations) fluids in circular tubes. The blood has viscosity value between 3 to 4 milli Pascal second (mPa s) or 0.3-0.4 millipoise at 37°C. Similarly food/liquid items that are used in your daily life such as water at 20 oC, milk at 25 oC, honey at 20°C, and ketchup at 20°C have viscosity values of 0.1, 0.3,

Aim: To determine viscosity co-efficient of a given liquid using Ostwald's viscometer

Principle: Viscosity arises due to internal friction between moving layers of molecules. A liquid flowing through a cylindrical tube of uniform diameter is expected to move in the form of molecular layers. A layer close to the surface is almost stationary while that at the axis of the tube moves faster than any other intermediate layer. A slow moving layer exerts a drag or friction on its nearest moving layer backwards. This property of a liquid by which it retards or opposes motion between layers is called viscosity. The co-efficient of viscosity is the tangential force per unit area required to maintain a unit velocity gradient between any two successive layers of a liquid situated unit distance apart. The coefficient of viscosity of a liquid is given by Poiseuille's formula.

In this experiment you will be determining the viscosity by studying the flow of fluid through a circular tube. Jean Poiseuille and Gotthilf Hagen first carried out these studies independently on various fluids. By studying the various factors that influence the flow of non-turbulent (steady) and non-pulsatile (no periodic variations) fluids through circular tube they arrived at an equation (1) given below for volume flow rate $\left(\frac{v}{t}\right)$[volume (v) that flows after a specified time (t)].

Volume flow rate $\left(\frac{v}{t}\right) = \frac{\pi \Delta P r^4}{8\,\eta l}$.....(1) alternatively, $\eta = \frac{\pi \Delta P\ t r^4}{8\ v l}$

Where v = volume of the liquid, r = radius of the tube, l = length of the tube, ΔP = pressure difference between the two ends of the tube, η = co-efficient of viscosity of the liquid.

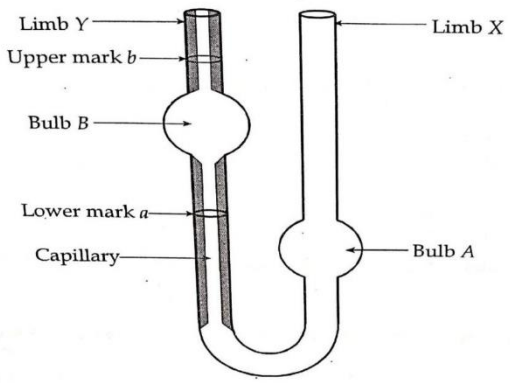
If equal volumes of two different liquids (one being water) are allowed to flow through the same tube under identical conditions, then

$$\frac{\eta l}{\eta w} = \frac{t l d l}{t w d w}$$

Procedure: Clean the given viscometer with water, rinse with acetone and dry it. Fix the viscometer vertically to a stand. Using a pipette, transfer a known volume of ion exchange water into the wider limb. Suck it above the upper mark of the viscometer. Allow it to flow freely through the capillary. When the level of water just crosses the upper mark, start a stop clock. Stop the stop clock when the water just crosses the lower mark. Record the time of flow in seconds. Repeat for 3 times.

Pour out the water, rinse the viscometer with acetone and dry it. Clamp it vertically to a stand and transfer the same volume of test liquid into the wider limb. As described above, record the time taken in seconds by the liquid to flow through tha same distance. Determine the coefficient of viscosity of liquid using the relation.

$$\frac{\eta liq}{\eta w} = \frac{t liq\ d liq}{t w\ d w}$$



Result:

The coefficient of viscosity of the given liquid = ... millipoise

Experiment: Observation and calculations

Liquid number:	Time of flow (in seconds)			
	Trail 1	Trail 2	Trail 3	Average
Test liquid (t_{liq})				
Water (t_w)				

Laboratory temperature =…….˚C (T)

Density of water at ……˚C = …….g/mL (will be provided to you)

Viscosity co-efficient of water at …. ˚C = …… millipoise (will be provided to you)

Density of the given liquid at ……. ˚C = …….g/mL (will be provided to you)

Viscosity co-efficient of the given liquid,

$$\eta_{liq} = \eta_w \frac{tliq\ dliq}{tw\ dw}$$

Where,

η_{liq}= coefficient of viscosity of the test liquid.

η_w= coefficient of viscosity of the water.

t_w= time taken (in seconds) by the water to flow from point A to B.

t_{liq}= time taken (in seconds) by the liquid to flow from point A to B.

d_{liq}= density of the given liquid.

d_w= density of water.

Result:

The coefficient of viscosity of the given liquid = …… millipoise

Model procedure

Experiment C2: Determination of Viscosity coefficient of lubricant (Ostwald's viscometer)

Pipette out 10 ml of the given liquid in to the wide limb of the dried viscometer and suck the liquid through the other limb. Determine the time of flow between two fixed points, one above and one below the bulb in the narrow limb of the viscometer. Repeat and calculate the average time of flow. Pour out the liquid, rinse the viscometer with Acetone and dry it. Now pipette out 10 ml of water into the wider limb and determine the average time of flow for water as before. From the density of the liquid and of water and the viscosity coefficient of water, determine the viscosity coefficient of the given liquid.

$$\eta_{liq} = \eta_w \frac{tliq\ dliq}{tw\ dw}$$

Where,

η_{liq}= coefficient of viscosity of the test liquid.

η_w= coefficient of viscosity of the water.

t_w= time taken (in seconds) by the water to flow from point A to B.

t_{liq}= time taken (in seconds) by the liquid to flow from point A to B.

d_{liq}= density of the given liquid.

d_w= density of water.

Experiment C3(CSE & ECE): Estimation of iron in TMT bar by diphenyl amine/external indicator method

Aim: To determine the percentage of iron in the TMT bar using standard $K_2Cr_2O_7$ solution.

Principle:

Iron (TMT) bar reacts with dil. HCl and forms its corresponding metal chloride salt ($FeCl_2$) and hydrogen gas. The balanced chemical reaction of dilute hydrochloric acid with TMT bar

$$Fe\,(s) + 2HCl\,(aq) \rightarrow FeCl_2\,(s) + H_2\,(g) \ldots\ldots\ldots\ldots (1)$$

In this experiment, a standard solution of potassium dichromate ($K_2Cr_2O_7$) is used to determine the percentage by weight of iron (as Fe^{2+}) in an unknown TMT bar sample. Dichromate ion reduces to two chromium (III) ions.
Therefore, 1 mole of $Cr_2O_7^{2-}$ (the oxidizing agent) reacts with 6 moles of Fe^{2+} (the reducing agent) to form 6 moles of Fe^{3+} and 2 moles of Cr^{3+}.
Thus, the overall redox reaction is:

$$Cr_2O_7^{2-} + 6Fe^{2+} + 14H^+ \rightarrow 6Fe^{3+} + 2Cr^{3+} + 7H_2O \ldots\ldots\ldots\ldots\ldots (2)$$

The reaction of Fe^{+2} with potassium ferricyanide $K_3[Fe(CN)_6]$ (use as an external indicator) results in the formation of a blue precipitate of ***Turnbull's blue*** (ferrous ferricyanide), but no blue colour formation with Fe^{+3} ion.

$$Fe^{+2} + K_3[Fe(CN)_6] \longleftrightarrow KFe[Fe(CN)_6] + 2K^+$$

Procedure:

Prepare 250 mL std $K_2Cr_2O_7$ solution by taking known wt of salt and dissolving in distilled water. Pipette out 25 ml of the $FeCl_2$ solution (TMT Bar) from the stock solution into a clean conical flask. Place a few numbers of drops of freshly prepared potassium ferricyanide indicator on wax paper. Add 1 mL of standard potassium dichromate from the burette to the conical flask containing $FeCl_2$ solution and mix well. Remove a drop of the solution from the conical flask and bring it in contact with a drop of the indicator on the wax paper. The colour of the indicator turns blue. Continue the titration by adding increments of 1ml of $K_2Cr_2O_7$ at a time and testing as above till a drop of the mixture fails to produce any colour with the indicator drop.

(Note: Clean the glass rod after every test). Repeat the titration by taking another 25 ml of ferrous chloride solution. This time add most of the potassium dichromate solution required at a stretch and then titrate drop-wise. Mix the flask's content after every addition and test a drop of the titrated solution with a drop of the indicator as described above till the indicator's color does not change.

Result:

The percentage of iron in the given iron solution taken from TMT bar is ……

Observation and Calculations

Part A: Preparation of standard solution of $K_2Cr_2O_7$.

1. Weight of bottle + $K_2Cr_2O_7$ salt = W_2= ---------- g

2. Weight of empty bottle = W_1 = ----------g

3. Weight of $K_2Cr_2O_7$ salt = W_2-W_1= ---------- g

$$Normality\ of \text{K2Cr2O7} = \frac{Weight\ of\ the\ salt\ (W2 - W1)\ X\ 4}{Eq.wt\ of\ \text{K2Cr2O7}(49\ g)} \quad \text{---- (`Z')}$$

Part B: Determination of percentage of Fe

Burette reading	Pilot reading	**Trial** I	**Trial** II
Final reading			
Initial reading			
Volume of $K_2Cr_2O_7$ run down (mL)			

Volume of $K_2Cr_2O_7$ consumed = ---------- mL (b)

Weight of TMT bar in 250 mL = ---------- g (W) ('W' will be provided to you).

1000 mL of 1N $K_2Cr_2O_7$ = 55.85 g of iron (1 equivalent of Fe = 55.85 g).

$$\text{`}b\text{'}\ \text{mL of `}a\text{'}\ \text{N}\ K_2Cr_2O_7 = \frac{55.85\ xaxb}{1000}\ \text{of Fe} = \text{---------- g } (c).$$

25 mL of sample solution contains = ---------- g of Fe (c).

250 mL of sample solution contains = 10 x c ---------- g of Fe (`d').

$$\text{The percentage of iron in given sample} = \frac{d\ X\ 100}{Weight\ of\ TMT\ bar\ (W)(g)}$$

Result:

The percentage of iron in the given iron solution taken from TMT bar is .....

Model procedure

Experiment C3. Estimation of iron in TMT bar by diphenyl amine/external indicator method

♦ **Part A**

Weigh out the given $K_2Cr_2O_7$ crystals accurately into a 250 ml volumetric flask, dissolve in distilled water and make up to the mark. Mix well and find the normality.

$$\text{Normality of } K_2Cr_2O_7 = \frac{\text{Weight of } K_2Cr_2O_7 \times 4}{\text{Equivalent weight of } K_2Cr_2O_7\ (49)}.$$

♦ **Part B**

Burette : Standard $K_2Cr_2O_7$ solution

Conical flask: 25ml of TMT bar solution

Indicator : Potassium ferricyanide $[K_3Fe(CN)_6]$ used as an external indicator

End point : No change in colour of indicator with the test solution drop

Conclusion : From the volume of $K_2Cr_2O_7$ consumed calculate the % of iron in TMT bar.

Conclusion: From the Volume of $K_2Cr_2O_7$ consumed, calculate the percentage of Iron in the given sample.

Experiment C4 (CSE & ECE): Estimation of Sodium present in soil/effluent sample using flame photometry

Significance of experiment

A photoelectric flame photometer is a device used in inorganic chemical analysis that uses the intensity of light emitted from a flame.[1] It has lots of applications.[2] It is used in Potash and fertilizer industry for highly accurate determination of sodium concentrations, Soil and environmental analysis for laboratory measurements for determination of alkali and alkaline earth elements, drinking water treatment: measurement of calcium and sodium concentrations in drinking water, glass industry for measurement of sodium concentration in glass and in clinical applications such as electrolyte determinations in blood and urin in areas without laboratory automation.

Aim: To estimate sodium from a soil/effluent sample solution using flame photometer.

Theory: Emission of characteristic radiation by an element and the correlation of the emission intensity with concentration of the element form the basis of flame photometry. When a solution containing sample element or ion is aspirated into the flame, following changes takes place, Firstly, solvent gets evaporated leaving behind salt in the flame. Then, salt gets evaporated into salt vapors, which further undergo dissociation into its constituent atoms. Some of the gaseous atoms formed may absorb heat energy from flame and get electronically excited to their higher energy level. Being unstable in the excited state, atoms fall back to their ground state, in form of light radiation.

Intensity of emitted light is proportional to number of atoms in the excited state, which in turn is proportional to the concentration of solution fed into the flame.

Different metals emit their characteristic radiations at different wavelength; they do not interfere with each other, even when they are present together.

Series of changes taking place at the flame are summarized as follows:

	Evaporation		Vaporization		Dissociation		
M+X-	→	M+X-	→	MX	→	MX	→ M (gas) + X (gas)
Solution		Mist		Solid		Gas	

Instrumentation: A flame photometer consists of an atomizer, mixing chamber, burner, filter, detector and a display device. Pressurized air is passed into atomizer and due to suction sample solution is drawn into the atomizer. Inside atomizer it mixes with air stream as a fine mist and passes into the mixing chamber, it mixes with gas and then passes into burner where mixture is burnt. The emitted radiation from flame passes through lens and then through a filter which allows only radiation characteristic of element under study to pass through detector. The output from detector is read out on a display device.

Procedure for determination of sodium: Transfer 2, 4, 6, 8 and 10 ml of standard sodium solution into different 25ml volumetric flasks from a burette. Make up all the solutions using distilled water and mix the solution well. To the given unknown soil/effluent solution add distilled water and shakes well. Switch on the instrument, turn the gas supply on and light the gas at the burner. Place the sodium filter (**589 nm**) in position. Now dip the capillary tube in a cell containing distilled water. Adjust the flame photometer to zero with respect to distilled water and hundred with respect to 100 ppm sodium solution.

Feed the various sodium solutions and test solution through the flame. Note down the flame photometer reading. Plot a graph of flame photometer readings against concentrations or volume of the solution to form the calibration curve. Using the curve obtained find out the volume of the unknown solution containing sodium ions and calculate the amount of sodium ions in it.

Result:

I) Concentration of sodium in the given unknown solution.....

II) The Amount of sodium in the unknown solution.....

Experiment: Observation and calculations

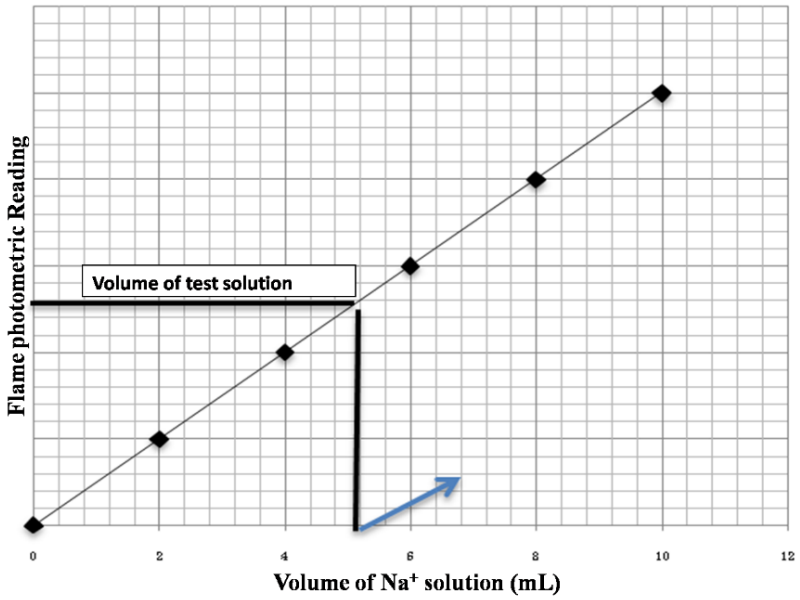
Concentration of solution: 1g of Na in 1000 mL of solution

Volume of sodium solution taken (mL)	Weight of sodium (g)	Concentration (ppm)	Flame Photometer reading
2			
4			
6			
8			
10			
Test			

Model procedure

Experiment C4. Estimation of Sodium present in soil/effluent sample using flame photometry

Transfer ***2, 4, 6, 8 and 10 ml*** of standard sodium solution into different ***25 ml*** volumetric flasks from a burette. Make up all the solutions including unknown soil/effluent solution using distilled water. After adjusting the air supply from the compressor, ignite the gas burner. Place the sodium filter (***589 nm***) in position and dip the capillary tube in a cell containing distilled water. The color of the flame completely turns to blue. Then, adjust the flame photometer reading to zero for ***calibration***. After calibration, feed the various sodium solutions prepared through the flame by spraying with atomizer one by one including the unknown solution. Plot a graph of flame photometer readings against volume of the Na^+ ion solution. From the curve find out the volume of the unknown solution containing sodium ions and calculate the amount of sodium ions in it.



Experiment D3(CSE): Design an experiment to identify the presence of proteins in given sample

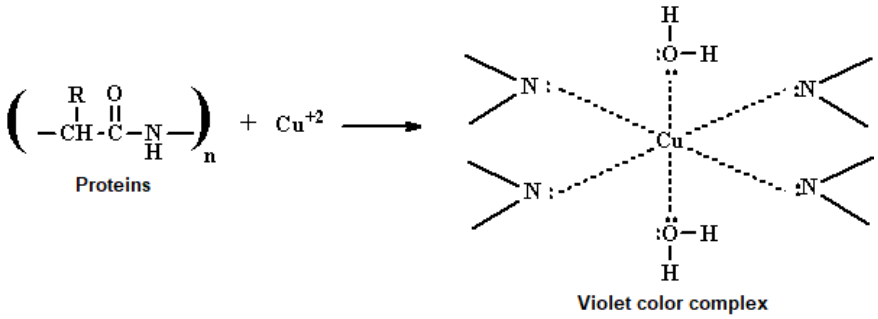
Significance

Proteins are a very versatile group of macromolecules and they form the fundamentals of critical processes in the body. At the cellular level, they control cell division, metabolism and the movement of a variety of material and information into and out of cells. These macromolecules ultimately have an impact on the structure, function and regulation of the tissues and organs in the body. So, it is important to elucidate the function of proteins and understanding the impact of their presence, absence and alteration. This is a key to advancing knowledge about diseases, providing the opportunity for biomarker discovery and development of therapeutics. Also the correct determination of the protein content of foods is important as, often, as is the case with milk, it determines the economic value of the food product and it can impact the economic feasibility of new industries for alternative protein production.

Aim: Identification of the presence of protein molecules via simple wet chemistry methods

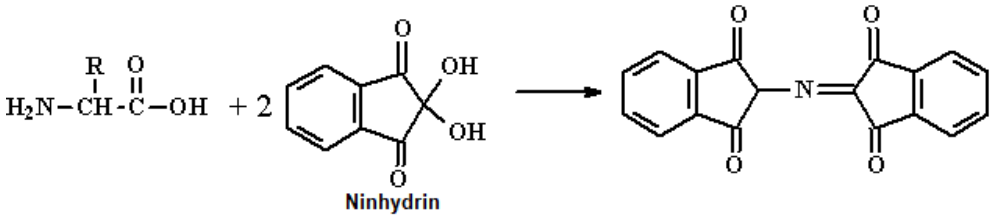
Theory: Protein has a high molecular mass long chain polymer composed of α-amino acids. Proteins are polypeptides of amino acids linked together by peptide bonds. Proteins are constituents of cells and hence present in living bodies. Proteins contain carbon, hydrogen, nitrogen, oxygen and sometimes phosphorus and sulphur. The following are the tests carried to find the presence of proteins in a given sample.

(a) Biuret Test: The compounds with peptide linkage undergoes this test. An alkaline solution of protein is treated with a drop of aqueous copper sulfate when a bluish violet color of complex is obtained. This test can be used for the identification of proteins and also for protein estimation.



(b) Xanthoproteic Test: Proteins on treatment with nitric acid gives a yellow or orange colour. Concentrated nitric acid is used for nitration. On the treatment of nitric acid, proteins give yellow precipitate which turns to orange colour on treatment with alkali.

(c) Ninhydrin Test: Proteins reacts with pyridine solution of ninhydrin gives coloured solution from deep blue to violet pink sometimes even red colour in some cases. Ninhydrin solution is prepared by dissolving 0.1gm of ninhydrin in about 100ml of distilled water. But this solution of ninhydrin is unstable and can be kept for two days.
The chemical reaction is given below.



Ninhydrin

Procedure

(a) Biuret Test:

Take the given sample to be tested in a clean test tube. Add 2ml of sodium hydroxide solution to it. To that add 5 to 6 drops of copper sulfate solution to it. The appearance of bluish violet colour indicates the presence of protein.

(b) Xanthoproteic Test:

Take 2ml of given sample compound in a test tube. Add a few drops of concentrated sulfuric acid and heat. If there is formation of yellow precipitate then the presence of protein is confirmed.

(c) Ninhydrin Test:

Take the sample solution to be tested in a clean test tube. Add 1-2ml of ninhydrin (0.2 grams of ninhydrin in 10ml of either ethanol or acetone) solution to it. Boil the mixture and observe the change. If there is the appearance of blue colouration then the presence of protein is confirmed.

Results

The given sample contains _____.

Experiment D4(CSE): Searching suitable PDB file and target for molecular docking

Aim: To have a basic idea about proteins and its structure. Searching suitable PDB file and target for molecular docking.

Protein

Proteins are large, complex molecules that are essential to the structure, function, and regulation of the body's cells, tissues, and organs. They are made up of long chains of smaller molecules called amino acids, which are linked together in a specific sequence via peptide bond to form a unique three-dimensional structure.

PDB file

The Protein Data Bank (PDB) is a public repository of three-dimensional structural data of biological macromolecules, such as proteins, nucleic acids, and complex assemblies. The PDB is managed and maintained by the Worldwide Protein Data Bank organization (wwPDB), which is a partnership among several organizations from around the world. The data in the PDB is obtained from experimental techniques such as X-ray crystallography, nuclear magnetic resonance (NMR) spectroscopy, and cryo-electron microscopy (cryo-EM).

The PDB provides access to the coordinates and other important data for more than 200000 macromolecular structures. This information is freely available to the scientific community, and it is widely used for a variety of purposes, including drug discovery, protein engineering, and molecular modeling. The PDB is constantly updated with new structures, and it is an essential resource for researchers in the field of structural biology

Contents of a PDB file

A PDB (Protein Data Bank) file is a file format used to store information about the three-dimensional structure of biological molecules, such as proteins, nucleic acids, and small molecules.

The contents of a PDB file typically include:

- **Header**: The header contains general information about the file, including the name of the molecule, the name of the authors who solved the structure, and the date of deposition.

- **Atom coordinates**: The coordinates of each atom in the molecule are listed in the file. These coordinates are typically derived from X-ray crystallography or NMR spectroscopy.
- **Connectivity information**: Information about the connectivity between atoms is also included in the PDB file. This information is used to build a three-dimensional model of the molecule.

- **Secondary structure information**: The PDB file may also contain information about the secondary structure of the molecule, such as alpha helices and beta sheets.
- **Chemical information**: The PDB file may contain information about the chemical composition of the molecule, including the names and types of amino acids or nucleotides.
- **Crystallographic information**: If the molecule was solved using X-ray crystallography, the PDB file may contain information about the crystal structure, such as unit cell dimensions and symmetry.

How to search a PDB file?

Search by PDB ID: The most straightforward way to search for a PDB file is to use the unique PDB ID assigned to each entry in the database. You can search for a PDB ID using the search bar on the PDB website (www.rcsb.org), or by including the PDB ID in your search query on other databases or search engines.

Instructions for PDB downloading

- -Type in name of protein (examples at bottom of the page).
- Click on Name icon (first name in purple box).
- -On the left side of the screen, click on Download/Display Structure
- -Under Download the Structure File, right click on the X where the PDB (top) meets with none, under compression (on left) in the table and save target as.
- Save under any filename you would like on your hard drive.

AIM of molecular docking

Molecular docking is a computational technique used to predict the binding orientation and affinity of a small molecule ligand to a target macromolecule, such as a protein, nucleic acid, or enzyme.

The goal of molecular docking is to predict the best conformation of a ligand molecule when it is bound to a receptor protein or other macromolecule, and to estimate the strength of the binding interaction between the two molecules. This information is important in drug

discovery, where the goal is to find small molecules that can bind selectively and tightly to a target protein or enzyme to modulate its activity.

There are many different software tools available for molecular docking, which use a variety of computational algorithms to predict ligand binding. Molecular docking is a useful tool for identifying new drug candidates, as well as for understanding the mechanisms of protein-ligand interactions in biological systems.

Result

Structure of Hemoglobin has been searched in PDB and is as follows.
(*Take a print out of Hemoglobin structure taken from PDB and paste it below*)

Experiment D3 (ECE): Determination of glucose by electrochemical Sensor

Significance

Glucose is the most important carbohydrate fuel in the body. In the fed state, the majority of circulating glucose comes from the diet; in the fasting state, gluconeogenesis and glycogenolysis maintain glucose concentrations. Very little glucose is found in the diet as glucose; most is found in more complex carbohydrates that are broken down to monosaccharides though the digestive process. About half of the total carbohydrates in the diet are in the form of polysaccharides and the remainder as simpler sugars. About two-thirds of the sugar in the diet is sucrose, which is a disaccharide of glucose and fructose. Glucose is classified as a monosaccharide because it cannot be broken down further by hydrolysis. It is further classified as a hexose because of its six-carbon skeleton and as an aldose, because of the presence of an aldehyde group on carbon 1. The aldehyde group condenses with a hydroxyl group so that glucose exists as a hemiacetal ring structure. This ring structure explains many of the reactions of glucose.Ordinarily the concentration of glucose in the blood is maintained at a relatively stable concentration from 80 to 120 mg/dl. The strong reducing properties of glucose made it relatively easy to measure and thus the clinical estimation of circulating glucose was one of the earliest tests available to the clinician. The recent introduction of microglucose oxidase technology has now made it possible for the patient to measure his or her own blood glucose concentration and undoubtedly makes the estimation of blood glucose the most widely used test of blood chemistry. An understanding of the methods of blood glucose measurement will help the clinician to interpret values and avoid the pitfalls of inaccurate testing.

Aim: To determine the level of glucose in blood samples

Working principle of glucose sensor The glucose sensor, determines the concentration of glucose in the solution. Most glucose meters, are based on electrochemical technology, they use electrochemical test strips to perform the measurement. In amperometric method, the electrochemical test strip contains an enzyme electrode containing an enzyme such as Glucose Oxidase. Glucose undergoes a chemical reaction (oxidation) in the presence of enzymes and electrons are produced during the chemical reaction, which leads to the generation of current. These electrons i.e., the charge passing through the electrode (Current) are measured which is proportional to the concentration of glucose in the solution.

Procedure:

Following steps are used to record the blood glucose level using glucometer

Step 1: Preparation of lancing device

- ✓ Prepare the lancing device by twisting and removing the mounting part.
- ✓ Internal cap is then re-attached to the mounting part of the device
- ✓ Select the depth of the blood sample between the range 1 and 5 where 5 means the deepest penetration

Step 2: Cleaning glucometer

Step 3: Preparation of glucometer and the strips

- ✓ Switch off the glucometer before inserting the sample sugar strips. Now insert the glucose test strip in the glucometer's test strip port. The glucometer will start automatically with a beep sound when the strip gets in its contact.

Step 4: Take the sample and monitor sugar level

- ✓ A blood sample is applied on the narrow end of the already inserted sugar testing strip.
- ✓ Wait for around 5 seconds to get the most accurate results. You will get the results given in g/l, mg/dl, or mmol/l.

Result

Concentration of sugar in the sample: 100 mg/dL

Experiment D4 (ECE): Synthesis of polyaniline and its conductivity measurement

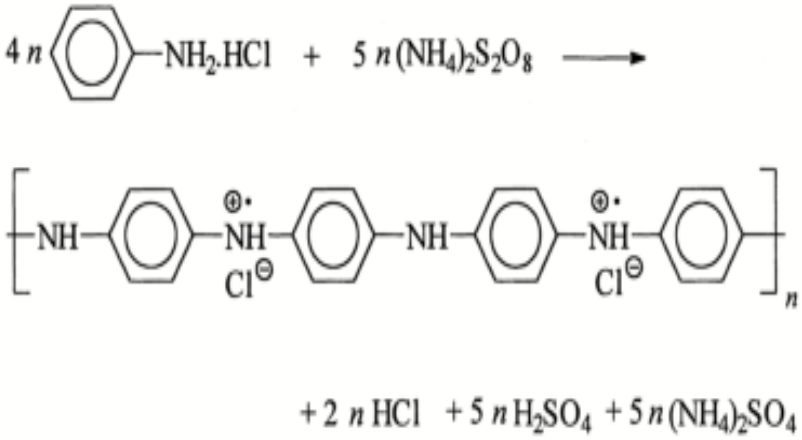
Significance of experiment:

The experiment will help to understand and familiarize with the synthesis process of polymeric materials, through the various aspects of reactants selection, stages of addition, washing and storage. PANI is a conducting polymer, so this is an application oriented synthesis process in which the product material will be characterized through its conductivity property. Conductivity (or specific conductance) of an electrolyte solution is a measure of its ability to conduct electricity. The SI unit of conductivity is siemens per meter (S/m). Conductivity measurements are used routinely in many industrial and environmental applications as a fast, inexpensive and reliable way of measuring the ionic content in a solution. For example, the measurement of product conductivity is a typical way to monitor and continuously trend the performance of water purification systems. Conductivity is traditionally determined by measuring the AC resistance of the solution between two electrodes. Dilute solutions follow Kohlrausch's Laws of concentration dependence and additivity of ionic contributions. Lars Onsager gave a theoretical explanation of Kohlrausch's law by extending Debye–Hückel theory.

Aim: Synthesis of polyaniline using ammonium peroxydisulfate oxidizing agent at room temperature and measurement of its electrical conductivity

Principle: The synthesis is based on mixing aqueous solutions of aniline hydrochloride and ammonium peroxydisulfate at room temperature, followed by the separation of PANI hydrochloride precipitate by filtration and drying. The goal of this study was to prepare PANI with a defined conductivity. The efficient polymerization of aniline is achieved only in an acidic medium, where aniline exists as an anilinium cation. For the present study, we have selected hydrochloric acid in equimolar proportion to aniline, i.e., aniline hydrochloride was used as a monomer. Peroxydisulfate is the most commonly used oxidant. The Polymerization is completed within 10 min at room temperature and within 1h at 0 –2 °C.

Conducting polymers like PANI has free electron in their matrix due to which they show electrical conductivity. Conducting polymers have been widely utilized in various applications due to their conductivity, compatibility and low-cost processability. In addition, compared with traditional metal or semiconductor materials, conducting polymers are more biocompatible. These advantages make conducting polymers more attractive to bioengineering researchers. The mechanism of electrical conductivity of conducting polymers is based on the transmission of polarons and bipolarons.



Procedure: PANI is chemically synthesized by oxidative polymerization. A monomer solution is prepared by adding 6 mL of aniline monomer into 7.5 mL of 1.5 M hydrochloric acid (HCl). Next, 0.75 g of ammonium persulfate (APS) powder is mixed with 10 mL of 1.5 M HCl solution to form the initiator solution. The mixture is stirred for 8 h at room temperature. The transformation of the solution to a blackish-green color indicates the formation of PANI emeraldine salt (PANI-ES). The addition of 0.1 M ammonium hydroxide (NH_4OH) into the PANI-ES solution formed PANI emeraldine base (PANI-EB). The obtained precipitate wash and dry to store.

To measure its electrical conductivity exactly 1g of synthesized PANI can be added into the given DI water taken in a clean 100 ml beaker and the electrode of the conductivity cell was immersed into the beaker and conductivity is measured using conductivity meter. Alternatively, make a coating of synthesized PANI on the electrode surface through drop casting using 1mg in 1ml solution of PANI from a micropipette and check the conductivity.

Result:

(i) Weight of polyaniline obtained =……..g
(ii) Color of polyaniline obtained =……..
(iii) Conductivity of PANI is =……..Ohm^{-1}

Links to the external sources of information about the topic:

doi:10.1088/1742-6596/1442/1/012003

doi:10.1088/1757-899X/772/1/012048

https://doi.org/10.1039/D1NJ03198H

http://en.wikipedia.org/wiki/Conductometry