STUDY MODULE- IV

Water Chemistry

(Physico-chemical Analysis of Water)

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List of Abbreviations

Sr. No.	Abbreviation	Expanded Form
1	EMR	Electromagnetic radiations
2	h	Planck’s constant
3	f	Frequency
4.	E	Energy
5.	V	Velocity
6.	m	meter
7.	nm	nanometer
8.	Hz	Hertz
9.	g	Grams
11.	mg/l	Milligrams per liter
12	ppm	Parts per million
13	L	liter
14	HCL	Hydrochloric acid
15	SMCL	Secondary maximum contaminant level
16	PSQCA	Pakistan Standards and Quality Control Authority
17	WHO	World health organization
18	SPADNS	Sulfanilic acid azochromotrop 2,7-Naphthalenedisulfonic acid, 4,5-dihydroxy-3-[(4-sulfophenyl)azo]-, trisodium salt
20	QC	Quality control
21	NSDWQ	Nigeria-Standard-for-Water-Quality
22	Conc.	Concentration
23	LED	light-emitting diode
25	ISE	Ion Selective Meter

Chapter # 1 UV-Vis Spectroscopy

1 Introduction of Spectroscopy

1.1 Definition

It is the branch of science that deals with the study of interaction of electromagnetic radiations with matter.

1.2 Electromagnetic Radiation EMR

It refers to the waves (or their quanta, photons) of the electromagnetic field, propagating (radiating) through space, carrying electromagnetic radiant energy. It includes radio waves, microwaves, infrared, (visible) light, ultraviolet, X-rays, and gamma rays.

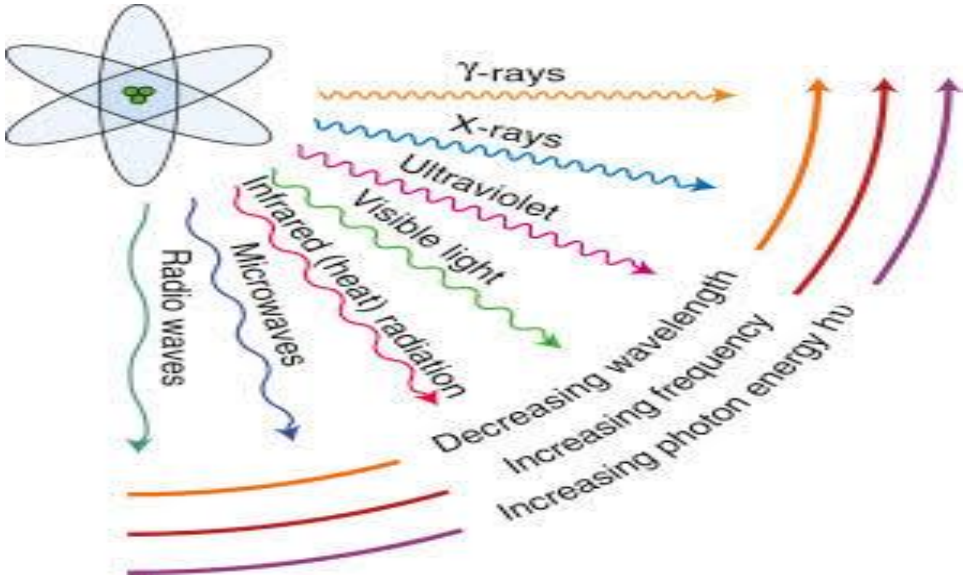

Figure 1: Types of Electromagnetic Radiations.

1.3 Energy of EMR

EMR is the type of energy that is transmitted through space at enormous velocities.

Energy is directly proportional to frequency.

$E = hf$

1.4 Spectrum

A spectrum is defined as the characteristic wavelengths of electromagnetic radiation (or a portion thereof) that is emitted or absorbed by an object or substance, atom, or molecule.

It is a graph of intensity of absorbed or emitted radiations by sample verses frequency or wavelength.

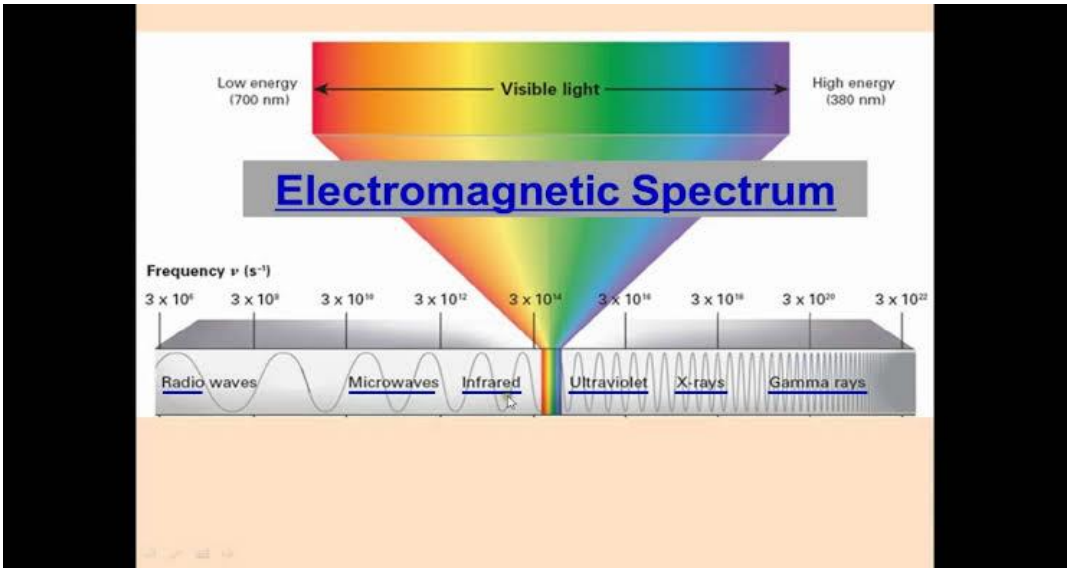

Figure 2: Spectrum of EMR

1.5 Frequency

It is the number of waves that pass a fixed place in a given amount of time.

Common symbols are f , v .SI unit is Hertz (Hz) A previous name for this unit was cycles per second (cps). The SI unit for period is the second.

Formula;

F = 1/T

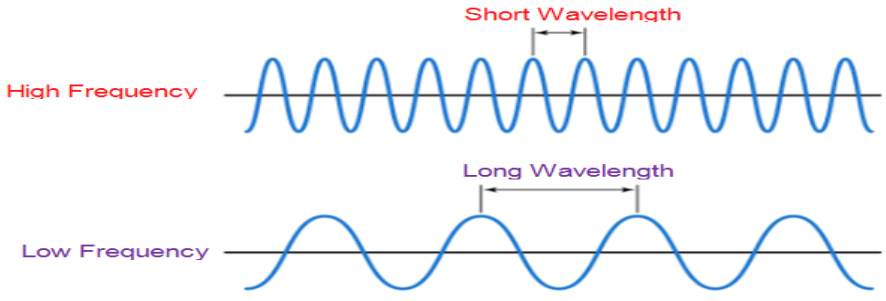

Figure 3: Frequencies of different radiations.

1.6 Wavelength

It is the distance between consecutive corresponding points of the same phase on the wave, such as two adjacent crests or troughs.

The lowercase version of the Greek letter "**lambda**" (**λ**) is the standard symbol used to represent wavelength.

Formula;

$\lambda = v/f.$

Unit; as **wavelength** is basically the distance, so the SI **unit** of **wavelength** is meters (m). Commonly wavelengths is written in nanometers (1 nm = 1e-9 m) or Ångström (1 å = 1e-10 m or 0.1 nm).

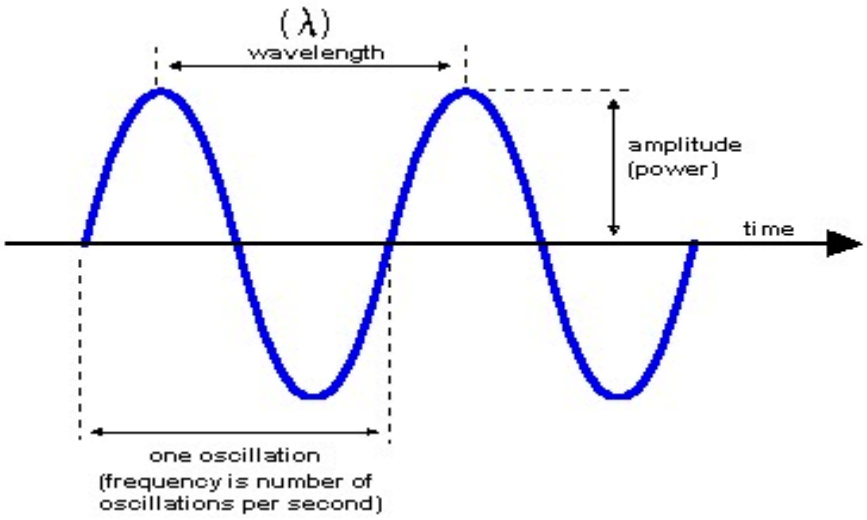

Figure 4: Wavelength

1.7 Types of Spectroscopy

There are many types of spectroscopy. Among these five types of spectroscopy are particularly useful to chemists.

There are infrared, ultraviolet/visible, nuclear magnetic resonance, atomic emission and absorption spectroscopy and mass spectroscopy.

1.7.1 How different types of radiations interact with chemicals

Sr.No	Types of radiations	Frequency range/Hz	Wavelength in nm	Effect on molecule	Types of spectroscopy
1	Ultraviolet	10^{15}-10^{17}	10 nm to 400 nm,	Excites the electron	Ultraviolet/visible spectroscopy
2	Visible light	10^{14}-10^{15}	400 nm to 750 nm	Excites the electron	Ultraviolet/visible spectroscopy

1.7.2 Electronic transitions in uv visible spectroscopy;

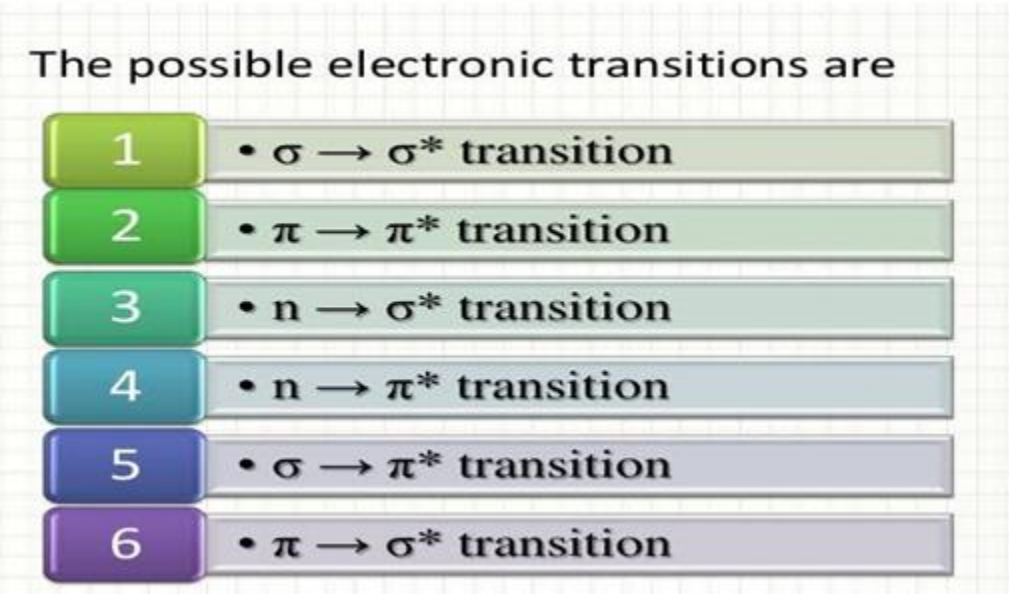

Figure 5: Different types of electronic transitions

1.8 Application of UV/VS Spectroscopy in Water Chemistry

It is routinely used in analytical chemistry for the quantitative determination of different analytes, such as fuctional groups, transition metal ions, highly conjugated organic compounds, and biological macromolecules.

1.9 Basic Principle of Spectroscopy

The principle is based on measurement of spectrum of a sample containing atoms/molecules. UV-Vis Spectroscopy is based on the Lambert-Beer principle which states that the;

Absorbance of a solution (A) is directly proportional to its pathlength (l) and its concentration (c) when the wavelength of the incidence light remains fixed. This is summarized in the following equation, where ε is the molar absorbtivity A = lc

1.10 Beer-Lambert Law

It states that the quantity of light absorbed by a substance dissolved in a fully transmitting solvent is directly proportional to the concentration of the substance and the path length of the light through the solution.

Mathematical statement of Beer's law is

A = εlc,

where: A = absorption; ε = molar attenuation coefficient, l = path length (the thickness of the solution), and c = concentration of the solution.

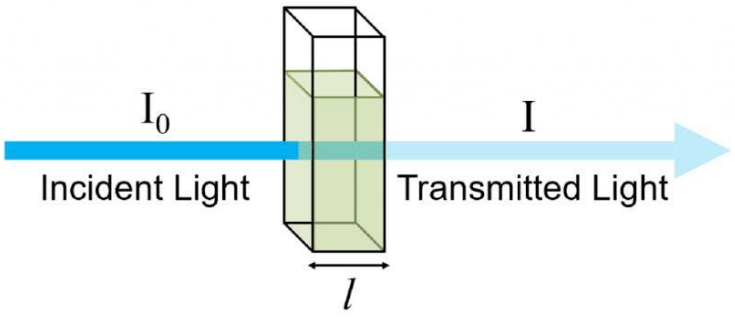

Figure 6: Incident Light versus Transmitted light

1.11 UV/VIS Spectrophotometer

The UV-Vis Spectrophotometer is the analytical instrument used for the UV-Vis spectroscopic analysis. Spectrophotometers are available in different configurations however most can be categorized into either single beam and double beam types depending on the design of their optical system.

1.12 Types of UV/VIS Spectrophotometer

1.12.1 Single Beam Spectrophotometer

A sample is placed in the UV/VIS beam and absorbance versus wavelength is measured. A single beam spectrophotometer utilizes one beam of light that passes through the sample and the intensity of the light reflected from a reference is measured without the sample.

1.12.2 Double Beam Spectrophotometer

The double beam approach to UV-Vis spectroscopy requires two beams of light, both having the same intensity to measure the Absorbance through sample and reference positions simultaneously. The Sample position is used for measurement of the analyte, whereas the reference position is used for the correction against a blank solution or sample matrix. A clear advantage of the double beam optical system is the improvement in measurement stability and drift precision as a result of having a real-time feedback of both the reference and sample signals.

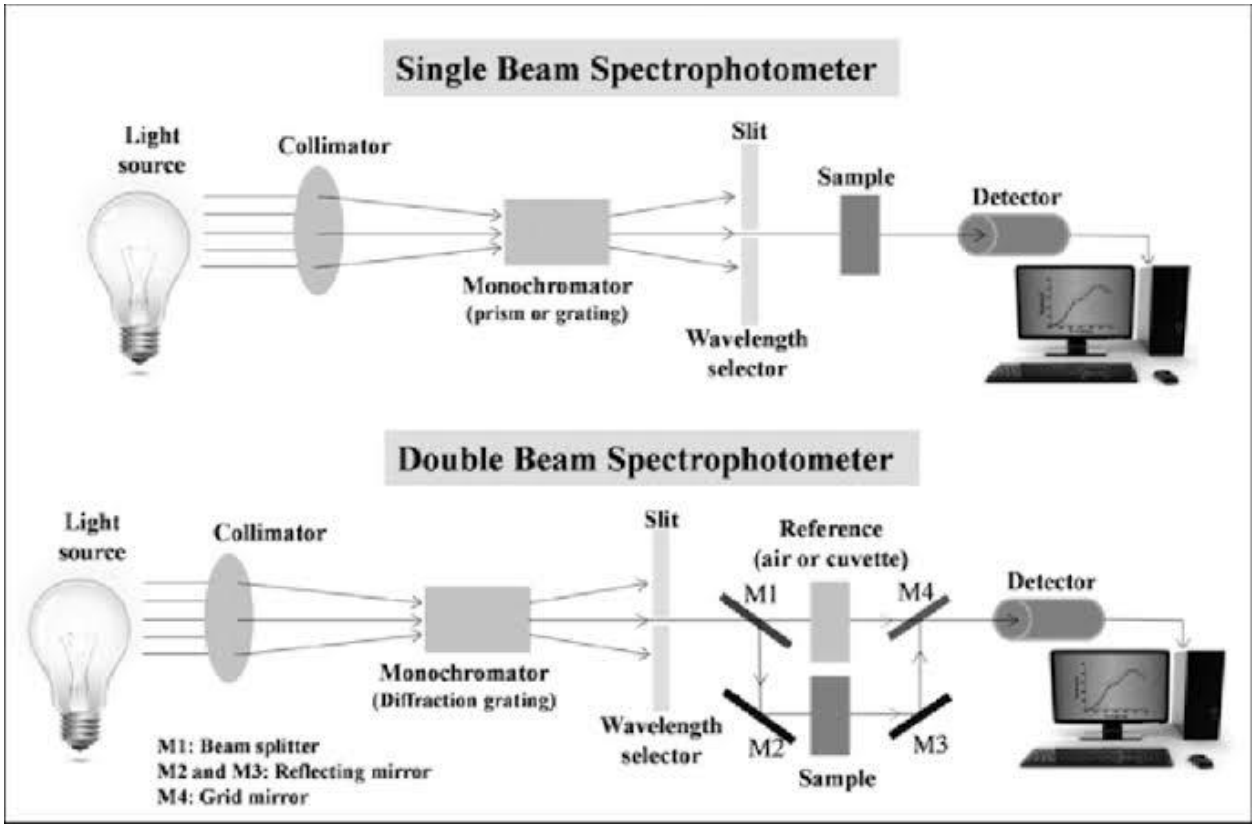

Figure 7: Difference between single and double beam UV/VIS Spectrophotometer.

1.13 Lambda max (λmax).

It refers to the wavelength along the absorption spectrum where a substance has its strongest photon absorption. It acts as a single quantitative parameter to compare the absorption range of different molecules.

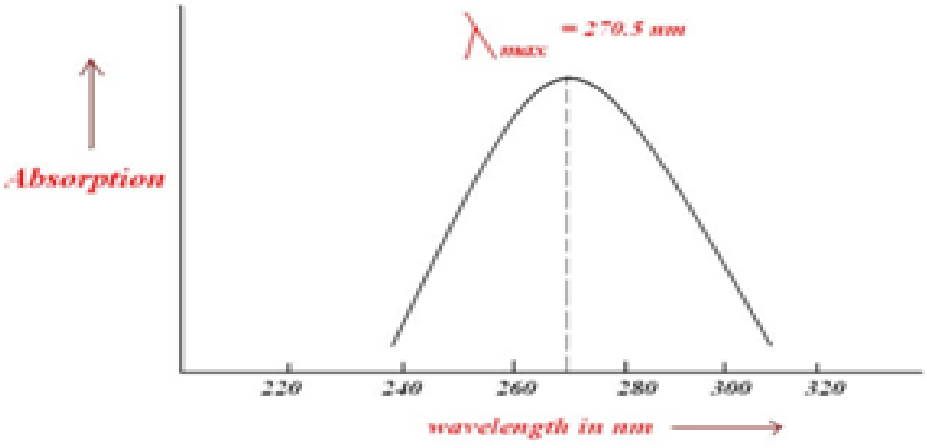

Figure 8: UV Spectra showing λmax of acetone

1.14 Learning Outcomes

On successful completion of this course students will be able to:

1 Explain fundamental concept of spectroscopy.

2 Recognize the common physical features of spectrophotometer.

3 Discuss beer lamberts law.

4 Illustrate the Electromagnetic radiations and electronic transitions responsible for the characteristic peaks and spectra lie under UV/VIS region.

1.15 Class Quiz

MCQs based questions have to be completed in limited time.

1.16 Group Exercise

Water Quality Assessment

Chapter # 2 UV-VIS Spectrophotometer

2 Introduction of UV-VIS Spectrophotometer

2.1 Definition

The scope of this equipment is the determination of different parameters such as Sulphate, Nitrate, Phosphate, Ammonia and Iron.

2.2 Testing Instrument

Model is UvLine- 9400 UV/VIS Spectrophotometer.

Figure 9: UvLine-9400 Spectrophotometer

2.3 Analytical Parameters

- Sulphate
- Nitrate
- Phosphate
- Ammonia
- Iron

2.4 Major Components

- Power supply
- Deuterium lamp
- Halogen lamp
- Quartz cells

2.5 Principle of Measurement

Light sources (Halogen & Deuterium lamp) produce the light that transmit or absorb the samples after being monochromatized in the spectrometer system. Photometer section split the light that exits the monochromater into sample and reference beam as well as shape the beam cross section in the sample compartment Equal optical path lengths and equal reflecting angles provides identical beam cross sections and polarization conditions in both beam paths.

2.6 Operation

- Turn on the power switch.
- Switch on the UVLINE-9400 by switching the power switch on the rear panel of the instrument.
- Depending on analytical requirement, either one or both light sources (Deuterium lamp, Halogen lamp) may be switched on. This is done during initialization of the instrument.
- After switching on the desired light source, activate parameter settings and select the relevant wavelength for the specific parameter.
- Also select the correction, cycle, and slit, Abs. /Trans. from the parameter setting window.
- Do special, wavelength and standard correction.
- Open the file of desired parameter, prepare standard curve by running the standards of different strengths and then directly measure the concentration of sample.

2.7 Quality Practices

2.7.1 Quality Control

- Spectrophotometer is standardized by running blank and known standard.
- Method blank is analyzed after every 10 samples.
- Laboratory Control samples are analyzed with every batch of 10 samples.
- NWQL check repeatability by reanalyzing a pre-analyzed sample once a day.
- To ensure the accuracy and precision of method spiking with known standard solution done once in month.

2.7.2 Cleaning

- Avoid keeping filled cells in the sample compartment longer than necessary to protect it against possible solvent vapors.
- Instantaneously wipe up samples spilt in the sample compartment or on accessory units using blotting paper.

- Wipe up any contamination on the instrument with a soft, clean cloth. If necessary, moisten the cloth slightly with, a commercial neutral detergent.

2.8 Maintenance

Equipment Preventive/Corrective actions taken are recorded in Equipment Maintenance log sheet.

2.8.1 Daily

- Carefully switch on and switch off the instrument.
- Avoid keeping filled cells in the sample compartment longer than necessary.
- Instantaneously wipe up samples spilt in the sample compartment or on accessory units.
- If high concentration of any chemical is kept for longer period then keep the sample cells dipped in 1:1HCL for 10-20 minutes before tap and deionized washing.
- Wash sample cells with deionized water after every sample analysis.
- Clean the instrument and its accessories with a soft, clean cloth by using a commercial neutral detergent.
- During cleaning keep the instrument switched off.
- Check voltage supply.
- Turn on the instrument and check for instrument accuracy by analyzing known standards.
- Dip reaction cells in (1:1) ethanol and diethyl ether for 24 hrs. for proper cleaning. Check under light that it should be properly cleaned.

2.8.2 Monthly

- Clean work place with a detergent.
- During cleaning keep the instrument switched off.
- The same procedure is adopted for instrument cleaning as in weekly maintenance.
- Check instrument alignment.
- Open the instrument cover to check the service life of deuterium lamp as its prolonged burning reduces its life.
- To open the instrument cover disconnects the power plug and switch off the instrument.

2.8.3 Yearly

- Unplug the instrument and clean the work place with a lab detergent.
- Avoid that no water enters in instrument.

- Switching the UVLINE-9400 frequently on/off reduces lamp life specifically this applies to deuterium lamp assuming its high standby voltage.
- Check lifetime of halogen and deuterium lamp assuming one switching operation the criterion for judging operating life of deuterium lamp is that its life may have drop to 50% after 1000 hrs.
- Replace the deuterium lamp if its energy is insufficient or the operating hours counter indicates 20(2000hrs).
- Replace halogen lamp also if it indicates energy insufficiency.
- To check fuses open the fuse holder by pulling its lid replace the defective fuses after unplugging the power cable.
- Carefully reassemble and reinitialize the instrument on a leveled surface free of vibrations.

2.9 Trouble Shooting

Sr. No.	Trouble Shooting (Display Shows)	Possible Causes	Remedies
1	Electric shock	Before opening of instrument it was not unplugged	Disconnect power plug before opening the instrument or removing any cover
2	Risk of burns	Halogen lamp has not cooled down	Prior to replacing bulb wait until they have sufficiently cooled down
3	Fire hazard	Protective cap of lamp was not removed	Remove the protected cap from the halogen lamp after its insertion to avoid fire hazards
4	Instrument not working	Check fuses of instruments	Only use fuse of the specified type and rating o Consider that their rating depends on line voltage

2.10 Precautions

- Connect the instrument to power outlet with earth conductor to ensure protection
- Maintain temperature range of +15…+350c.
- Keep the instrument at vibration and `dust free place.

- There should be no splashing of water near instrument.
- Never look directly in to the lamp radiation.

2.11 Planned Demonstration

Operation of UV/VIS Spectrophotometer in Chemical Lab III (quality control, maintenance, calculations).

2.12 Learning Outcomes

On successful completion of this course students will be able to:

1 Explain principle of measurement.

3 Recognize the physical operation of UV/VIS Spectrophotometer.

4 Illustrate the special, wavelength and standard correction procedure and open the file of desired parameter, preparation of standard curve by running the standards of different strengths.

5 Discuss the quality control practices and periodic maintenance of UV/VIS Spectrophotometer.

2.13 Class Quiz

2.14 Group Exercise

Water Quality Assessment

Chapter # 3 Nitrate

3 Introduction of Nitrate

3.1 Introduction

Water is a precious commodity and is in limited amount. Nearly 70 % of the earth's surface is covered by water, which is an important source for us. Only 2.66% of the total global water resources including groundwater, lakes, rivers, polar ice and glaciers are freshwater. A small fraction of freshwater (0.6% is usable as drinking water. Therefore, water resources must be managed properly and treatment of wastewater must be done efficiently. Rapid population growth, agriculture industries, and domestic waste have resulted into elevated level of pollutants. Continuously increasing rate of population is affecting the quality of water and ultimately converting it into wastewater.

Nitrate is the major nutrient needed by living microorganism for their physiological processes. However they are considered as pollutants if their concentration is more than recommended limit. High concentration of nitrate favors the growth of aquatic plants but create negative effect on water quality by accelerating the growth of algal clump, bad odor, and discoloration. Such conditions create problems in its use for recreational and aesthetic purposes. Excessive growth of aquatic life causes problems in navigation and aeration.

3.1.1 Sources of Nitrate in Water

Nitrate can get into water directly as the result of runoff of fertilizers containing nitrate. Ammonia and organic nitrogen can enter water through sewage effluent and runoff from land where manure has been applied or stored.

Main sources are;

- nitrogenous fertilizers,
- municipal wastewater,
- septic system leakage,
- urban drainage reject,
- refuse dumps,
- sewage discharge,
- industrial discharge,
- contaminated land,
- unhygienic sanitation practices,
- landfills, animal waste, and soluble nitrogen compounds contained by geologic materials—aggravate the nitrate contamination

- The urine and excreta of farm animals such as cows and buffaloes and poor casings have also been linked to nitrate contamination in shallow groundwater.

3.1.2 Importance of Nitrate in Water

Nitrogen is essential for all living things as it is a component of protein. Nitrogen exists in the environment in many forms and changes forms as it moves through the nitrogen cycle. Nitrogen is the essential component of agricultural soil. Nitrate is used by plants and animals and eventually returns to the air as nitrogen gas. In nature, plants utilize nitrate as an essential nutrient. In commerce, the majority of nitrate is used in inorganic fertilizers. Urea, ammonium nitrate, calcium nitrate, potassium nitrate, and ammonium phosphate are some example of nitrogen fertilizers, easily soluble in water and are used extensively in the preparation of single-nutrient or multinutrient fertilizer solutions.

3.1.3 Impacts

3.1.3.1 Health Impacts (Water borne Diseases caused by high nitrate in water)

Toxicity of nitrates causes methemoglobinemia, carcinogenicity, goiter, birth defects, abortions, histopathological changes in cardiac muscles, diabetes mellitus, and livestock poisoning. Nitrate ingestion may lead to oxygen deprivation in infants of below 6 months of age causing methemoglobinemia or blue baby syndrome. Approximately 20% of the nitrate secreted in saliva is converted to nitrite by the microorganisms present in the oral cavity in individuals with high rate of conversion in contrast to normal individuals with 5% conversion. The radical conversion of nitrate to nitrite post ingestion forming carcinogenic N-nitrosamines on reaction with amines and amides can lead to potential cancer risk. Nitrates can also cause interference to the normal iodine metabolism of the thyroid gland.

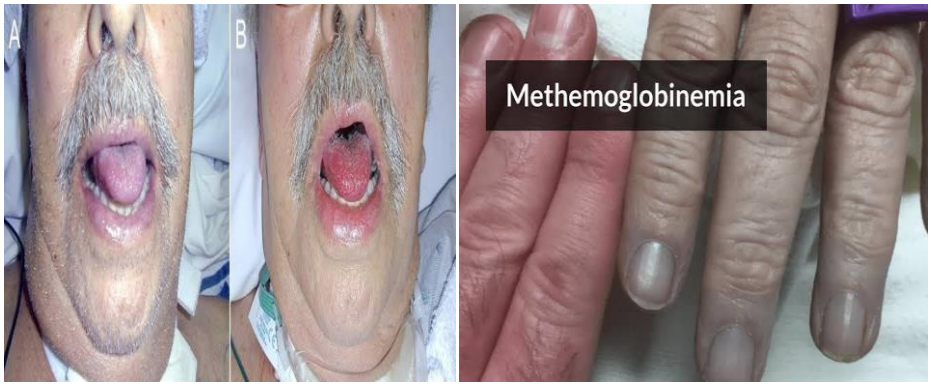

Figure 10: Methemoglobinemia caused by high nitrate in drinking water.

3.1.3.2 Environmental Impacts

Excess levels of nitrates in water can create conditions that make it difficult for aquatic insects or fish to survive.

Algae and other plants use nitrates as a source of food. If algae have an unlimited source of nitrates, their growth is unchecked.

Eutrophication – "The process by which a body of water acquires a high concentration of nutrients, especially phosphates and nitrates. These typically promote excessive growth of algae. As the algae die and decompose, high levels of organic matter and the decomposing organisms deplete the water of available oxygen, causing the death of other organisms, such as fish.

Anoxia – Anoxic Event: Anoxia is a lack of oxygen caused by excessive nutrients in waterways which triggers algae growth. When the plants die and decay, oxygen is stripped from the water, which then turns green or milky white and gives off a strong rotten egg odour. The lack of oxygen is often deadly for invertebrates, fish and shellfish.

Excess plants and algae will also create conditions where organic matter accumulates. High densities of algae will create a condition where sunlight cannot reach very far into the water. Since plants and algae require some sunlight, plants and algae not receiving sunlight will die off. These dead plant materials will settle to the bottom of the water and bacteria that feed on decaying organic material will greatly increase in numbers. These bacteria will consume oxygen and, therefore, the level of dissolved oxygen in this water will fall to levels that are too low for many aquatic insects and fish to survive. Also, this can cause extreme changes in habitat. Fish that need gravel or sand for spawning may find nothing but mats of vegetation and muck so will be unable to produce offspring.

3.1.4 Permissible Limits

- The Environmental Protection Agency (EPA) has since adopted the 10 mg/L standard as the maximum contaminant level (MCL) for nitrate-nitrogen in drinking water. Permissible limit by PSQCA/NSDWQ, 2010 is 10ppm or 10 mg/l.
- According to Bottled water standards (PSQCA 2010): Permissible limit is 10mg/l.

3.1.5 Nitrate Treatment Methods

Nitrate may be successfully removed from water using treatment processes such as ion exchange, distillation, and reverse osmosis. Heating or boiling water will not remove nitrate. Nitrate testing kits are also available at domestic level.

3.2 Determination of Nitrate (NO_3^{-1}) by Spectrophotometer

3.2.1 Definition

The purpose of this method is to characterize the quality of potable water. Knowledge of Nitrate allows a true picture of water quality and clear indication of sewerage contamination.

3.2.2 Testing Instrument UVLine -9400 UV/VIS Spectrophotometer

3.2.3 Analytical Parameters

- Phosphate
- Nitrate
- Nitrite
- Silica
- Iron
- Sulphate

3.2.4 Major Components

- Power supply
- Deutrium lamp
- Halogen lamp
- Quartz cells

3.2.5 Principle of Measurement

Technique is useful for screening sample that have low organic content. Measurement of UV absorption at 220nm enables rapid determination of nitrate while UV absorption at 275nm give indication of organic interferences UV screening technique on spectrophotometer follow Beer's Lambert Law.

3.2.6 Standards and Reagents

a) Preparation of 100mg/l Nitrate ($NO3^{-1}$) Stock Solution

Reagents

- Potassium Nitrate
- Distilled water
- Chloroform

Procedure

- Weigh 0.7218g well-dried KNO3.

- Quantitatively transferred to 1000 ml Volumetric Flask.
- Make the volume up to the mark with distilled water.
- Transfer to an appropriately label bottle container.
- Preserve stock solution by adding 2 ml of chloroform in 1L (the solution is stable for at least six months).
- Transfer to an appropriately labeled bottle container with sticker.

b) Preparation of Nitrate $(NO_3)^{-1}$ Working Standards

Reagents

- 100 mg/l Potassium Nitrate stock
- Distilled water

Procedure

- Take 10 ml of 100-mg/l stock solution in to 100 ml to prepare a 10 mg/l secondary standard.
- Make the volume up to the mark with distilled water.
- Verify standards with previously made standards by comparing the two curves.
- Prepare standards of 1,2---7 ppm using formula C1V1=C2V2 by making dilution from 100 mg/l Nitrate stock solution.
- Transfer to an appropriately labeled bottle container with sticker.

3.2.7 Calibration of Spectrophotometer for Nitrate

- Prepare Nitrate standards from 1 to 7 ppm (1,2,3,4,5,6,7) from 100 ppm potassium nitrate stock solution.
- Now set the spectrophotometer according to equipment instructions by applying wavelength and standard correction.
- Take distilled water as blank and consider all standards as samples.
- Take 10 ml sample (Standard) + 0.2 ml 1N HCL. Shake well and take absorbance reading at wavelength of 220 nm or construct calibration curve.
- Repeat the procedure with the same sample at wavelength of 275 nm.
- Using corrected standards absorbances obtain sample concentration by comparing absorbance or directly from Standard curve.

3.2.8 Testing

- Take 10ml deionized water in beaker and add 0.2 ml of 1N HCL in it.
- Apply special correction or blank correction.
- Now take 10ml standard or sample in cuvet and add HCL to it.

- Take absorbance reading at 220nm to measure nitrate concentration in the sample and at 275nm to determine organic interference.
- Subtract two times absorbance reading at 275nm from the reading at 220nm to obtain corrected reading.
- Determine the concentration from equation.
- Conc. of sample= $\frac{\text{Abs. of sample x Conc. of standard}}{\text{Abs. of standard.}}$
- If Spectrophotometer is set for concentration determination, directly measure concentration from calibration curve.

3.2.9 Quality Control

- Method blank is analyzed after every 10 samples.
- Laboratory Control samples of 05 ppm analyzed with every batch of 10 samples.
- NWQL check repeatability by reanalyzing a pre-analyzed sample once a day.
- To ensure the accuracy and precision of method spiking with known standard solution done once in month.
- Results of laboratory control sample are regularly displayed in the form of control charts.

3.2.10 Expression of Results

The nitrate is measured in mg/l.

3.2.11 Interpretation of Results

Compare the results with Water Quality Standards or Guidelines to predict the safe or unsafe level.

3.2.12 Precautions

- Wash the reaction cell properly with distilled water before analysis.
- Apply Reagents blank correction and standard adjust features to compensate for variations in reagents.
- If correction value is more than 10% of the reading at 220 nm, use other method for nitrate analysis.
- The method gives best result in range of 1-7 mg/l nitrate. For higher concentration do dilutions.

3.2.13 Planned Demonstration

Testing of Nitrate in Chemical Lab III (Reagents preparation, glassware washing, standards preparation, testing, quality control, calculations and reporting, unit's conversion).

3.2.14 Learning Outcomes

1 Explain the importance of nitrate in water.

2 Identify the basic sources and impacts of high concentrations of nitrate in water.

Prepare the reagents and standards to construct the calibration curve.

3 Illustrate the procedure to calibrate the instrument for the determination of nitrate.

4 Perform the testing of nitrate from water samples.

5 Discuss water quality data, safe and unsafe levels.

On successful completion of this course students will be able to:

3.2.15 Class Quiz

3.2.16 Group Exercise

Water Quality Assessment

Chapter# 4 Sulphate

4 Introduction of Sulphate

4.1 Introduction

Sulfate is second to bicarbonate as the major anion in hard water reservoirs. Sulfate is one of the major dissolved components of rain. High concentrations of sulfate in the water we drink can have a laxative effect when combined with calcium and magnesium; these are the two most common constituents of hardness.

4.1.1 Sources in Water

Sulfates occur naturally in numerous minerals, including barite (BaSO4), epsomite ($MgSO_4 \cdot 7H_2O$) and gypsum (CaSO4·2H2O). These dissolved minerals contribute to the mineral content of many drinking-waters. Sulfates are discharged into water from mines and smelters and from kraft pulp and paper mills, textile mills and tanneries. Sodium, potassium and magnesium sulfates are all highly soluble in water, whereas calcium and barium sulfates and many heavy metal sulfates are less soluble. Atmospheric sulfur dioxide, formed by the combustion of fossil fuels and in metallurgical roasting processes, may contribute to the sulfate content of surface waters. Sulfur trioxide, produced by the photolytic or catalytic oxidation of sulfur dioxide, combines with water vapour to form dilute sulfuric acid, which falls as “acid rain”.

4.1.2 Importance of Sulphate in Water

Inorganic sulfate (SO_4^{2-}) is required for the synthesis of 3′-phosphoadenosine-5′-phosphosulfate (PAPS). PAPS are required for synthesis of many important sulfur-containing compounds, such as chondroitin sulfate and cerebroside sulfate. Chondroitin Sulfate has several functions. It delivers nutrients to the joint cartilage, helps to inhibit the enzymes that decompose the joint cartilage and speeds up the formation of a new joint cartilage. Cerebrosides are the common name for a group of glycosphingolipids called monoglycosylceramides which are important components in animal muscle and nerve cell membranes. While significant levels of sulfate are found in foods and various sources of drinking water, the major source of inorganic sulfate for humans is from biodegradation due to body protein turnover of the sulfur amino acids methionine and cysteine. Dietary sulfate in food and water, together with sulfate derived from methionine and cysteine found in dietary protein and the cysteine component of glutathione, provides sulfate for use in PAPS biosynthesis. Sulfate requirements are thus met when intakes include recommended levels of sulfur amino acids.

4.1.3 Impacts

4.1.3.1 Health Impacts

Bacteria that live in soil use sulfur as a food or energy source. The bacteria then produce hydrogen sulfide gas. When you smell the odor of sulfur in your water, that lets you know that you're ingesting the waste of bacteria. High concentration of sulfur in drinking water can lead to diarrhea and dehydration.

4.1.3.2 Environmental Impacts

Sulfur not only stinks and makes water taste bad; it can also stain sinks, toilets, and clothing and even damage plumbing. The bacteria that create the sulfur smell produce a slime that can potentially corrode the plumbing pipes.

Figure 11: Staining of tap due to high concentration of Sulphur in water.

4.1.4 Permissible Limits

- Sulfate is classified under the secondary maximum contaminant level (SMCL) standards. According to PSQCA, 2010 the SMCL for sulfate in drinking water is 250 milligrams per liter (mg/l), sometimes expressed as 250 parts per million (ppm).
- According to Bottled water standards (PSQCA 2010): Permissible limit is 250mg/l.

4.1.5 Sulphate Treatment Methods

Several methods of removing sulfate from water are available. The treatment method selected depends on many factors including the level of sulfate in the water, the amount of iron and manganese in the water, and if bacterial contamination also must be treated. The option you choose also depends on how much water you need to treat.

For treating small quantities of water (drinking and cooking only) the typical methods may be distillation or reverse osmosis. The most common method of treating large quantities of water is ion exchange. This process works similar to a water softener. Ion-exchange resin, contained inside the unit, adsorbs sulfate. When the resin is loaded to full capacity with sulfate, treatment ceases. The resin then must be "regenerated" with a salt (sodium chloride) brine solution before further treatment can occur.

Distillation boils water to form steam that is then cooled and then recondense the water. Minerals, such as sulfate, do not vaporize with the steam and are left behind in the boiling chamber. *Reverse osmosis* membranes have a porosity that permits water molecules to pass through but leaves the large ions in solution.

4.2 Determination of Sulphate (SO_4^{-2}) by Spectrophotometer

4.2.1 Definition

- The purpose of this method is to characterize the quality of potable water in context to Sulphate estimation in water.

4.2.2 Testing instrument

UvLine 9400 UV/VIS Spectrophotometer.

4.2.3 Analytical Parameters

- Phosphate
- Nitrate
- Nitrite
- Silica
- Iron
- Sulphate

4.2.4 Major Components

- Power supply
- Deutrium lamp
- Halogen lamp
- Quartz cells

4.2.5 Principle of Measurement

Sulphate ion (SO_4^{-2}) is precipitated in an acidic (pH 2.50-3.50) medium with barium chloride ($BaCl_2$) so as to form barium sulphate crystals of uniform size. Measurement as absorbance of the barium sulphate suspension on spectrophotometer is based on Beer's Lambert Law.

4.2.6 Standards and Reagents

Standards

- Sulphate Buffer
- Barium Chloride Crystals (Commercially available)
- Distilled water

a) Preparation of 1000 mg/l Stock Solution Sulphate

Reagents

- Na_2SO_4 (anhydrous)
- Distilled Water

Procedure

- Weigh 1.4791 gm of anhydrous Na_2SO_4.
- Quantitatively transfer to 1000 ml volumetric flask.
- Dilute to mark with distilled water.
- Transfer to a bottle container labeled with sticker.

b) Preparation of 100ppm $(SO_4)^{-2}$ Solution & Sulphate Working Standards

Reagents

- 1000 mg/l SO_4 Stock Solution
- Distilled Water

Procedure

- Transfer 100 ml of 1000-mg/l stock solution in to 1000 ml volumetric flask to produce a 100 mg/l secondary standard.
- Make to volume with distilled water.
- Check standards accuracy on instrument against previous standards.
- Prepare standards of 5, 10, 15---40 ppm using formula $C_1V_1=C_2V_2$ by making dilutions from 100 ppm Sulphate standard in 100 ml Volumetric flasks
- Transfers to an appropriately labeled container with label**.**

c) **Preparation of 10ppm Sulphate (SO_4)-2 Standard (lab control sample)**

Reagents

- 100 mg/l SO_4 Solution
- Distilled Water

Procedure

- Transfer 10 ml of 100 mg/l stock solution in to 100 ml volumetric flask to produce a 10 mg/l secondary standard.

- Make to volume with distilled water.
- Transfer to an appropriately labeled container with label.

d) Preparation of Buffer Solution for Sulphate (SO_4)$^{-2}$

Reagents

- Magnesium Chloride
- Sodium Acetate
- Potassium Nitrate
- Sodium Sulphate
- Acetic Acid
- Distilled water

Procedure

e) Preparation of Buffer Solution-A

- Weigh 30g $MgCl_2.6H_2O$ in to 100 ml beaker.
- Weigh 5g $CH_3COONa.3H_2O$ and 1.0g KNO_3.
- Dissolve all the three reagents in 500 ml distilled water and add 20 ml CH_3COOH (99%) to it.
- Now make volume up to the mark with distilled water and shake well.

f) Preparation of Buffer Solution-B

- When sample sulphate concentration is less than 10 mg/L, 0.111g sodium sulphate is added before addition of acetic acid after which volume is made up to 1000 ml.
- Transfer to a 1000 ml bottle container and label with Sticker.

4.2.7 Calibration of Spectrophotometer for Sulphate

- Prepare Sulphate standards from 5 to 40 ppm (5,10,15,20,25,30,35,40) from 100 ppm sodium sulphate stock solution.
- Now set the spectrophotometer according to equipment instructions by applying wavelength and standard correction.
- Take distilled water as blank and consider all standards as samples.
- Take 10 ml sample + 2 ml buffer (Use Buffer solution B for construction of calibration curve of 5 & 10 ppm standards and Buffer solution A for calibration curve of 10-40 ppm standards) +a pinch of barium chloride and take absorbance after 5 minutes reaction time at wavelength of 420 nm or directly construct calibration curve.

4.2.8 Calibration of Spectrophotometer for Sulphate

- Prepare Sulphate standards from 5 to 40 ppm (5,10,15,20,25,30,35,40) from 100 ppm sodium sulphate stock solution.
- Now set the spectrophotometer according to equipment instructions by applying wavelength and standard correction.
- Take distilled water as blank and consider all standards as samples.
- Take 10 ml sample + 2 ml buffer (Use Buffer solution B for construction of calibration curve of 5 & 10 ppm standards and Buffer solution A for calibration curve of 10-40 ppm standards) +a pinch of barium chloride and take absorbance after 5 minutes reaction time at wavelength of 420 nm or directly construct calibration curve.

4.2.9 Testing

- Take 10ml of deionized water in cuvet and add 2 ml of Sulphate Buffer (Use Buffer solution B for samples having concentration >10ppm and Buffer solution A for other).
- Add one pinch of Barium chloride crystals in the above solution, stir it vigorously and take absorbance reading after 5 minutes reaction time at wavelength of 420nm.
- Now perform with actual water samples and determine the concentration from equation
- $\text{Conc. of sample} = \frac{\text{Abs.of sample x Conc.of standard}}{\text{Abs.of standard}}$
- If Spectrophotometer is set for concentration determination, directly measure concentration from calibration curve.

4.2.10 Quality Control

- Method blank is analyzed after every 10 samples.
- Laboratory Control samples of 10 ppm analyzed with every batch of 10 samples.
- NWQL check repeatability by reanalyzing a pre-analyzed sample once a day.
- To ensure the accuracy and precision of method spiking with known standard solution done once in month.
- Results of laboratory control sample are regularly displayed in the form of control charts.

4.2.11 Expression of Results

The Sulphate is measured in mg/l.

4.2.12 Interpretation of Results

Compare the results with Water Quality Standards or Guidelines to predict the safe or unsafe level.

4.2.13 Precautions

- Wash the cuvets properly with distilled water before analysis.
- Apply Reagents blank correction and standard adjust features to compensate for variations in reagents.
- The pH of the buffer solution should be adjusted at 2.50-3.50 range to avoid decomposition of barium sulphate.
- Above 40 ppm accuracy of curve decreases therefore it is recommended to make dilutions if the concentration of sample is more than 40 ppm.

4.2.14 Planned Demonstration

Testing of sulphate in Chemical Lab III (Reagents preparation, glassware washing, standards preparation, testing, quality control, calculations and reporting, unit's conversion).

4.2.15 Learning Outcomes

On successful completion of this course students will be able to:

1. Explain the importance of sulphate in water.

2. Identify the basic sources and impacts of high concentrations of sulphate in water.

3. Prepare the buffer solutions and working standards to construct the calibration curve.

4. Illustrate the procedure to calibrate the instrument for the determination of sulphate.

5. Perform the testing of sulphate from water samples.

6. Discuss water quality data, safe and unsafe levels.

4.2.16 Class Quiz

4.2.17 Group Exercise

Water Quality Assessment

4.3 Alternate Method for the Determination of Sulphate by Colorimeter

4.3.1 Definition

The purpose of this method is to characterize the quality of potable water in context to sulphate estimation in water.

4.3.2 Testing Instrument Colorimeter Model DR/890 Hach

Figure 12: HACH colorimeter model DR/890.

4.3.3 Analytical Parameters

- Phosphate
- Nitrate
- Nitrite
- Silica
- Iron
- Sulphate

4.3.4 Major Components

- Sample Cells
- COD Adopter

- Batteries (4) AA Alkaline
- Key Pad

4.3.5 Principle of Measurement

The HACH DR/890 series colorimeter is a microprocessor controlled LED-sourced filter photometer suitable for colorimetric testing in the laboratory or field. The instrument uses visible light for measurement of different parameters.

4.3.6 Standards and Reagents

Process of Standard and reagents preparation is the same as mentioned in above method (spectrophotometric method).

4.3.7 Calibration of Colorimeter for Sulphate

- Using colorimeter press.
- EXIT.
- SETUP then press⇩ 2times.
- Press ENTER and enter program 101 then ENTER.
- Press⇩ one time then press ENTER.
- Enter 1 and ENTER.
- Press⇩ for absorbance mode.
- Then apply blank sample. Take 10 ml deionozed water; add 2ml buffer and pinch of barium chloride.
- Cap the sample cell and give 5 minutes reaction time.
- Shake the sample cell vigorously.
- Place the sample cell into cell holder of colorimeter. Tight the instrument cap.
- Time beeps, press zero.
- Place the standards with same procedure and press read.
- If instrument give exact reading to standards proceed with sample analysis.

4.3.8 Testing

- Take 10ml of deionized water in cuvet and add 2 ml of Sulphate Buffer (Use Buffer solution B for samples having concentration >10ppm and Buffer solution A for other).
- Add one pinch of Barium chloride crystals in the above solution, stir it vigorously and take absorbance reading after 5 minutes reaction time at wavelength of 420nm.
- Now perform with actual water samples and determine the concentration from equation
    - $\text{Conc. of sample} = \frac{\text{Abs.of sample x Conc.of standard}}{\text{Abs.of standard}}$

4.3.9 Quality Control

- Colorimeter is standardized by running blank and known standard.
- Method blank is analyzed after every 10 samples.
- Laboratory Control samples are analyzed with every batch of 10 samples.
- NWQL check repeatability by reanalyzing a pre-analyzed sample once a day.
- Daily quality control data is recorded in QC-data sheet -1.

4.3.10 Expression of Results

The Sulphate is measured in mg/l.

4.3.11 Interpretation of Results

Compare the results with Water Quality Standards or Guidelines to predict the safe or unsafe level.

4.3.12 Precautions

- Wash the sample cells properly.
- Acid washing is necessary when going to another parameter to avoid interferences of reagents and instrument accuracy.
- After adding the reagents, shake properly for the specific time.
- Regularly check instrument batteries.

4.3.13 Planned Demonstration

Testing of Sulphate in Chemical Lab III (Reagents preparation, glassware washing, standards preparation, testing, quality control, calculations and reporting, unit's conversion).

4.3.14 Learning Outcomes

On successful completion of this course students will be able to:

1. Illustrate the procedure to calibrate the instrument for the determination of sulphate.

2. Perform the testing of sulphate from water samples.

3. Discuss water quality data, safe and unsafe levels.

4.3.15 Class Quiz

4.3.16 Group Exercise

Water Quality Assessment

Chapter#5 Flouride

5 Introduction of Flouride

5.1 Introduction

Fluorine is a common element that does not occur in the elemental state in nature because of its high reactivity. It accounts for about 0.3 g/kg of the Earth's crust and exists in the form of fluorides in a number of minerals.

5.1.1 Sources in Water

Sources of fluoride contamination in drinking water are both natural and anthropogenic activities. Marine aerosols, volcanic eruptions, some geothermal activities and fluoride bearing rock's minerals are mainly natural sources of fluoride in environment. Apart from these natural sources, there are some anthropogenic sources also that increase the fluoride concentration in environment. Industrial aerosols and phosphate fertilizers plants, sewage sledges and pesticides are main anthropogenic sources for high concentration of fluoride in soil and water. There are some other minor factors also that affect the fluoride level in groundwater such as temperature, pH of water and soil, soil's sorption capacity, depth of well and climatic conditions of any area.

5.1.2 Importance of Fluoride in Water

The presence of environmental fluoride and its impact on human health is well documented. When consumed in adequate quantity, fluoride prevents dental caries, assists in the formation of dental enamels, and prevents deficiencies in bone mineralization.

5.1.3 Impacts

5.1.3.1 Health Impacts

Its excessive intake may result in slow, progressive crippling scourge known as fluorosis (dental as well as skeletal). Excessive consumption of fluoride may lead to muscle fibre degeneration, low haemoglobinlevels, deformities in RBCs, excessive thirst, headache, skin rashes, nervousness, neurological manifestations (it affects brain tissue similar to the pathological changes found in humans with Alzheimer's disease), depression, gastrointestinal problems, urinary tract malfunctioning, nausea, abdominal pain, tingling sensation in fingers and toes, reduced immunity, repeated abortions or still births, male sterility, etc. It is also responsible for alterations in the functional mechanisms of liver, kidney, digestive system, respiratory system, excretory system, central nervous system and reproductive system, destruction of about 60 enzymes. The effects of fluoride in drinking water on animals are analogous to those on human beings.

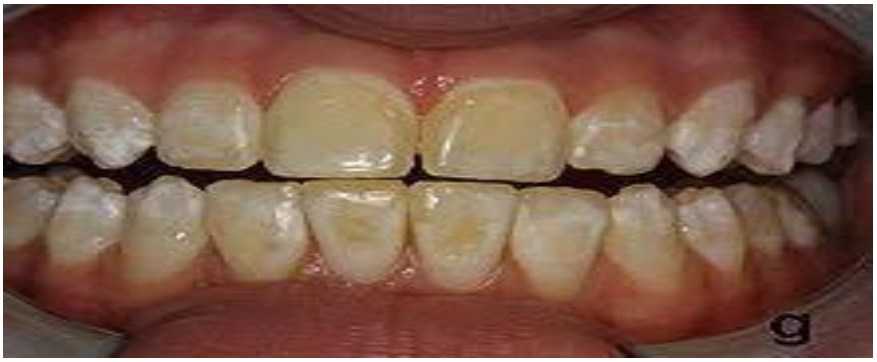

Figure 13: Dental flourosis

5.1.3.2 Environmental Impacts

Signs of harmful effects on plants include the yellowing of leaves and slowed growth. When plants take up fluoride from soil its toxic effects depend on the ionic species of fluoride present and on the type of soil. Many studies have assessed the effects of fluoride emissions on plants by exposing them to hydrogen fluoride (HF) gas. Studies shows that the smaller the distance from great fluoride sources such as aluminium smlters or phosphorus plants, the higher the fluoride levels in soils and hence the degree of damage to vegetation.

Harmful effects of fluoride, including species vulnerability, have been reported in an array of wild animals. Even domestic pets have been subjects of reports raising concerns about fluoride exposure, especially through their water and food. Additionally, the effects of fluoride on farm animals have been documented. Health problems include anorexia, cramping, collapse, respiratory and cardiac failure, and death.

5.1.4 Permissible Limit

- PSQCA/NSDWQ, 2010 recommended limit is 1.5 mg/l or ppm.
- According to Bottled water standards (PSQCA 2010): Permissible limit is 0.7mg/l.

5.1.5 Fluoride Treatment Methods

There are several methods which are conventional treatment methods like adsorption, ion exchange, reverse osmosis, electrodialysis, coagulation and precipitation etc can be practiced at community level or at households to reduce fluoride concentration before ingestion. But the choice of each method depends on the local conditions of the region such as the quality of groundwater and the source of contamination whether it is natural or anthropogenic. Fluoride contamination being a prominent and widespread problem in several parts of the world and as causes for this are mostly natural and unpreventable, educating the people and defluorinating the groundwater before consumption are essential for a healthy world.

5.2 Determination of Fluoride by ISE Meter

5.2.1 Definition

The purpose of this method is to characterize the quality of potable water in context to fluoride estimation in water.

5.2.2 **Testing Instrument** Ion meter Model JENWAY 3345

Figure 14: ISE meter of JENWAY

5.2.3 Analytical Parameters

- Fluoride
- Chloride
- Iodide

5.2.4 Major Components

- Base block
- Swing arm
- Integral temperature probe
- Ion selective electrode

5.2.5 Principle of Measurement

ISE meter working is based on the type of electrode used for ion analysis. The fluoride electrode is an ion-selective sensor. The key element in the fluoride electrode is the laser-type doped lanthanum fluoride crystal across which a potential is established by fluoride solutions of different concentrations. The crystal contacts the sample solution at one face and an internal reference solution at the other. The fluoride electrode measures the ion activity rather than concentration.

5.2.6 Standards and Reagents

a) Preparation of 100 mg/l Fluoride $(F)^{-1}$ Stock Solution

Reagents

- Sodium Fluoride NaF (Anhydrous).
- Distilled water.

Procedure

- Weigh 221.0 mg anhydrous sodium Fluoride.
- Quantitatively transfer to 1000 ml volumetric flask.
- Dilute to mark with distilled water.
- Transfer to an appropriately labeled bottle container with label.

b) Preparation of Fluoride Working Standards.

Reagents

- 100 mg/l Sodium Fluoride NaF.
- Distilled water.

Procedure

- Transfer 10 ml of 100 mg/l stock solution into 100 ml volumetric flask to produce a 10 mg/l secondary standard.
- Make the volume up to the mark with distilled water.
- Dilute to mark with distilled water.
- Prepare standards of 0.2 and 2 ppm using formula $C_1V_1=C_2V_2$ by making dilutions from 10 ppm Fluoride standard.
- Transfer to appropriately labeled containers with sticker.

c) Preparation of Ion Strength Adjuster for ISE Meter

Reagents

- Sodium Fluoride
- Glacial Acetic Acid
- Sodium Hydroxide
- Distilled Water

Procedure

- Weigh 58 gm NaF.
- Dissolve in 500 ml distilled water.

- Add 57 ml of Glacial Acetic Acid in 500 ml of NaF solution.
- Add 4.0 g 1,2 Cyclohexylene diamine tetra-acetic acid.
- Adjust the pH of solution using 5M NaOH solution (pH 5-5.5).
- After maintaining the pH of solution makes the volume up to the 1000 ml mark.
- Transfer to a bottle container and label with sticker.

5.2.7 Calibration of ISE Meter for Fluoride

- Connect the fluoride electrode to the instrument.
- Prepare standards of 0.2, 2 ppm from 10 ppm fluoride stock solution.
- Take 10 ml of standard and add 10 ml of fluoride buffer to it. Note the reading on instrument while continuously stirring the cuvet.
- If equipment does not give exact reading of standard press “calibrate.” Instrument will get automatically calibrated and will give exact value of standard. Now proceed with samples.

5.2.8 Testing

- Take 10 ml deionized water in 50 ml cuvet and add 10 ml fluoride Buffer (pH: 5.3-5.5) in it.
- Dip the electrode in the solution with constant stirring.
- This will show zero concentration. Now similarly note the concentration of known standards for verification of calibration and efficiency of electrode.
- Then take 10 ml sample and analyze it as described above.
- Numbers of replicates to be analyzed are “Three” for each sample.
- Use samples and standards at the same temperature.

5.2.9 Quality Control

- Method blank is analyzed after every 10 samples.
- Laboratory Control samples of 5ppm analyzed with every batch of 10 samples.
- NWQL check repeatability by reanalyzing a pre-analyzed sample once a day.
- To ensure the accuracy and precision of method spiking with known standard solution done once in month.
- Daily quality control data is recorded in QC Data sheet-I.
- Results of laboratory control sample are regularly displayed in the form.

5.2.10 Expression of Results

The Fluoride is measured in mg/l.

5.2.11 Interpretation of Results

Compare the results with Water Quality Standards or Guidelines to predict the safe or unsafe level.

5.2.12 Precautions

- First check the reliability of the instrument by running known standards.
- The pH of the buffer solution must be in range of 5.3-5.5 for the accurate work.
- Alkalinity, iron, aluminum and free chlorine may interfere during the analysis of fluoride.
- It should be kept in mind that at alkaline pH the hydroxide ions also interfere the electrode response.

5.2.13 Planned Demonstration

Testing of Fluoride in Chemical Lab III (Reagents preparation, glassware washing, standards preparation, testing, quality control, calculations and reporting, unit's conversion).

5.2.14 Learning Outcomes

On successful completion of this course students will be able to:

1. Explain the importance of fluoride in water.
2. Identify the basic sources and impacts of high concentrations of fluoride in water.
3. Prepare the reagents and working standards.
4. Illustrate the procedure to calibrate the ISE meter for the determination of fluoride.
5. Perform the testing of fluoride from water samples.
6. Discuss water quality data, safe and unsafe levels.

5.2.15 Class Quiz

5.2.16 Group Exercise

Water Quality Assessment

5.3 Alternate Method for the Determination of Fluoride (F^-) by SPADNS Method

5.3.1 Definition

The purpose of this method is to characterize the quality of potable water in context to fluoride estimation in water.

5.3.2 Testing Instrument DR/2800Colorimeter, HACH

Figure 15: DR/2800Colorimeter, HACH

5.3.3 Analytical Parameters

- Fluoride
- Iron

5.3.4 Major Components

- Protective cover
- Source Lamp Tungsten
- square matched glass sample cells
- Cell adapters

5.3.5 Principle of Measurement

The SPANDS colorimetric method is based on the reaction between fluoride and a zirconium-dye lake. Fluoride reacts with the dye lake, dissociating a portion of it into a colorless complex anion (ZrF_6^{-2}) and the dye. As the amount of fluoride increases, the color becomes progressively lighter.

5.3.6 Standards and Reagents

- SPANDS solution (Commercially Available)
- Distilled Water

5.3.7 Calibration of colorimeter DR/2800 for Fluoride

- Turn on the power switch.
- Keep the lid covered. System will start the self check for system check, lamp test, filter adjustment, wavelength adjustment and voltage test.
- Select the stored program for fluoride.
- Press the start button.
- Prepare standard of 1ppm from 100ppm working standard.
- Apply the blank sample by deionized water 10 ml and 2 ml SPADNS solution in 25 ml curette give one minute reaction time by pressing timer, press zero then press read.
- Take 10 ml of standard solution and 2 ml of SPADNS Solution in 25 ml curette. Give one minute reaction time. After the timer beeps press read.

5.3.8 Testing

- Take 10ml deionized water in 25 ml cuvet and add 2 ml of SPANDS solution in it.
- Shake well and give one minute reaction time.
- Make blank correction, then take 10ml sample and treat it as described above and measure concentration.
- Number of replicates to be analyzed are “Three” for each sample.

5.3.9 Quality Control

- Method blank is analyzed after every 10 samples.
- Laboratory Control samples of 1 ppm analyzed with every batch of 10 samples.
- NWQL check repeatability by reanalyzing a pre-analyzed sample once a day.
- To ensure the accuracy and precision of method spiking with known standard solution done once in month.
- Daily quality control data is recorded in QC Data sheet-I.
- Results of laboratory control sample are regularly displayed in the form of control charts.

5.3.10 Expression of Results

The fluoride is measured in mg/l.

5.3.11 Interpretation of Results

Compare the results with water quality standards or guidelines to predict the safe an unsafe level.

5.3.12 Precautions

- Check the calibration of instrument before and after use.
- Take accurate volume of SPANDS and sample because it is extremely important for analytical accuracy.
- Use samples and standards at the same temperature or at least within 2 °C difference.

5.3.13 Planned Demonstration

Testing of fluoride in Chemical Lab III (Reagents preparation, glassware washing, standards preparation, testing, quality control, calculations and reporting, unit's conversion).

5.3.14 Learning Outcomes

On successful completion of this course students will be able to:

1. Explain the importance of fluoride in water.
2. Identify the basic sources and impacts of high concentrations of fluoride in water.
3. Prepare the reagents and working standards.
4. Illustrate the procedure to calibrate the colorimeter for the determination of fluoride.
5. Perform the testing of fluoride from water samples.
6. Discuss water quality data, safe and unsafe levels.

5.3.15 Class Quiz

5.3.16 Group Exercise

Water Quality Assessment

Chapter#6 Iron

6 Introduction of Iron

6.1 Introduction

Iron is the second most abundant metal in the earth's crust, of which it accounts for about 5%. Elemental iron is rarely found in nature, as the iron ions Fe2+ and Fe3+ readily combine with oxygen- and sulfur-containing compounds to form oxides, hydroxides, carbonates, and sulfides.

6.1.1 Sources of Iron

It is one of the earth’s most plentiful resources. Rainwater as it infiltrates the soil and underlying geologic formations dissolves iron, causing it to seep into aquifers that serve as sources of groundwater for wells. The source of iron may be from the corrosion of iron or steel pipes or other components of the plumbing system where the acidity of the water, measured as pH, is below 6.5. Although present in drinking water, iron is seldom found at concentrations greater than 10 milligrams per liter (mg/L) or 10 parts per million. However, as little as 0.3 mg/l can cause water to turn a reddish brown color. Iron is not hazardous to health, but it is considered a secondary or aesthetic contaminant. Essential for good health, iron helps transport oxygen in the blood. Most tap water in the United States supplies approximately 5 percent of the dietary requirement for iron.

6.1.2 Importance of Iron

Essential for good health, iron helps transport oxygen in the blood. Most tap water in the United States supplies approximately 5 percent of the dietary requirement for iron. Iron is an important component of hemoglobin, the substance in red blood cells that carries oxygen from your lungs to transport it throughout your body. If you don't have enough iron, your body can't make enough healthy oxygen-carrying red blood cells. A lack of red blood cells is called iron deficiency anemia. Iron is involved when a plant produces chlorophyll, which gives the plant oxygen as well as its healthy green color. This is why plants with an iron deficiency, or chlorosis, show a sickly yellow color to their leaves. Iron is also necessary for some enzyme functions in many plants.

6.1.3 Impacts

6.1.3.1 Health Impacts

Iron in water is considered as a contaminant because it also contains bacteria that feed off it. In addition to this, high iron content leads to an overload Iron overload is caused by a mutation in the gene that digests iron; this mutation affects around one million people in the United States which can cause diabetes, hemochromatosis, stomach

problems, nausea, and vomiting liver, heart and pancreatic damage, as well as diabetes. Early symptoms include fatigue, weight loss, and joint pain.

Water with excessive amounts of dissolved minerals such as iron and magnesium can have negative effects on your skin. They can damage healthy skin cells, which can lead to wrinkles.

In addition, water with iron doesn't blend well with soap. This causes issues when showering and bathing, as soap scum residue will be left not only in your bathtub but on your skin as well. This can clog skin pores, which will lead to a buildup of oils in your skin. This, in turn, can lead to skin problems such as acne or eczema.

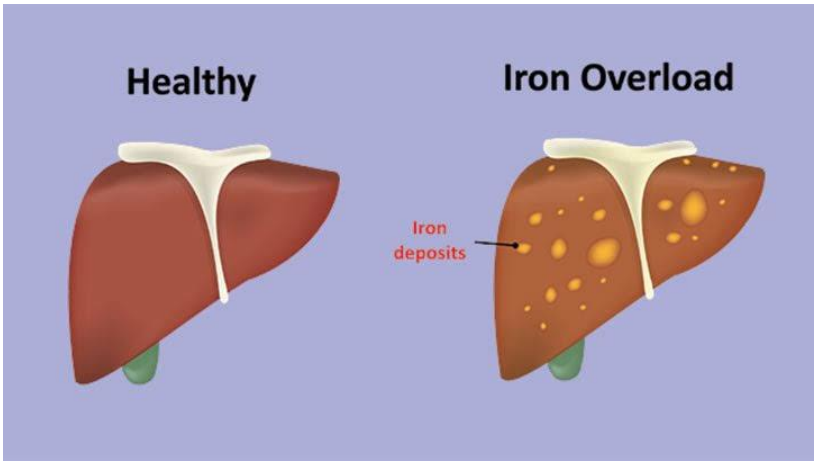

Figure 16: Hemochromatosis

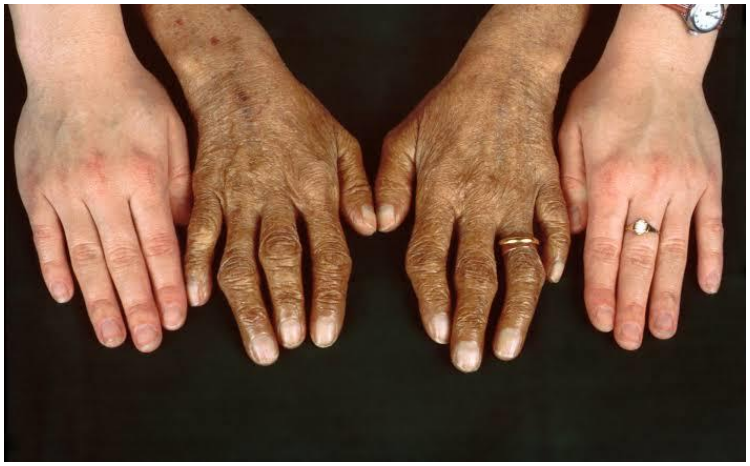

Figure 17: Iron damages the healthy skin and leads to wrinkles.

6.1.3.2 Environmental Impacts

Taste and Food

Dissolved ferrous iron gives water a disagreeable metallic taste. When the iron combines with tea, coffee and other beverages, it produces an inky, black appearance and a harsh, unacceptable taste. Vegetables cooked in water containing excessive iron turn dark and look unappealing.

Stains and Deposits

Concentrations of iron as low as 0.3 mg/L will leave reddish brown stains on fixtures, tableware and laundry that is very hard to remove. When these deposits break loose from water piping, rusty water will flow through the faucet.

Iron Bacteria

When iron exists along with certain kinds of bacteria, a smelly bio film can form. To survive, the bacteria use the iron, leaving behind a reddish brown or yellow slime that can clog plumbing and cause an offensive odor. This slime or sludge is noticeable in the toilet tank when the lid is removed. The organisms occur naturally in shallow soils and groundwater, and they may be introduced into a well or water system when it is constructed or repaired.

6.1.4 Permissible Limits

- WHO, 2004 recommended the limit for iron is 0.3 mg/l or ppm.
- According to Bottled water standards (PSQCA 2010): Permissible limit is 0.3mg/l.

6.1.5 Iron Treatment Methods

If there is an iron problem with the water supply, the first step is to determine the source. The source of iron may be from the corrosion of iron or steel pipes or other components of the plumbing system where the acidity of the water, measured as pH, is below 6.5.

Table 1: Treatment Methods for Various Forms of Iron

Symptoms	Form of Iron	Treatment Methods	Considerations
Tap water is first clear and colorless. After standing, reddish brown particles	Dissolved ferrous iron	Aeration/Filtration	Temperature dependent
		Water softener	Hardness must be calculated and increased sodium concentration should be checked if users(s) on restricted sodium diet. System

appear and settle to bottom of glass.			must be airtight.
		Chlorination/Filtration	Use of chlorine liquid or pellets. Requires frequent monitoring and proper water pressure. May require lengthy contact time.
		Manganese Greensand/Filtration [1]	Adequate pressure
		Catalytic filtration[2]	Dissolved oxygen, alkalinity, organic matter, chlorination, polyphosphate, and temperature limitations
		Ozonation	Cost
		Sequestering (adding chemical agents to water to keep iron to an insoluble, filterable form)	Method may not prevent staining and may require removal of sequestering agents and iron. Test for agents before choosing another treatment device.
Tap water appears rusty or has a red or yellow color. After standing, particles settle to bottom.	Insoluble red water ferric iron	Manganese Greensand/Filtration[1]	Adequate pressure
		Catalytic filtration[2]	Dissolved oxygen, alkalinity, organic matter, chlorination, polyphosphate, and temperature limitations
		Chlorination/Filtration	Use of chlorine liquid or pellets. Requires frequent monitoring and proper water pressure.
Water tank, toilet tank and plumbing have reddish brown	Iron bacteria	Shock chlorination; consider following with continuous	Chlorine products must be suitable for drinking water. Method requires long contact

or yellow gelatinous slime or sludge present. Odor may be objectionable.		chlorination.	time for adequate treatment.
Water containing organic iron is usually yellow or brown color, but may be colorless. Tannins stain water a tea color.	Organic iron and tannins[3]	Water softener	First, treat for organics (activated carbon). Check for corrosive properties. System must be airtight.
		Manganese Greensand/Filtration[1]	First, treat for organics. Maintain adequate pressure.
		Ozonation	Cost

1. Manganese Greensand: A naturally occurring mineral or manufactured material, treated with potassium permanganate that is capable of removing iron; it absorbs dissolved iron and requires chemical regeneration.
2. Catalytic Filtration: A granular filter medium that enhances the reaction between oxygen and iron and then filters the insoluble iron.
3. Since organic iron and tannins can slow or prevent iron oxidation, water softeners, aeration systems, and iron filters may not work satisfactorily. One option may be chemical oxidation followed by filtration.

6.2 Determination of Iron (II) by Spectrophotometer

6.2.1 Definition

The purpose of this method is to characterize the quality of potable water in context to iron estimation in water.

6.2.2 Testing Instrument

Model UvLine 9400 UV/VIS Spectrophotometer

6.2.3 Analytical Parameters

- Iron
- Nitrate
- Sulphate
- ammonia

6.2.4 Major Components

- Power supply
- Deuterium lamp
- Halogen lamp
- Quartz cells

6.2.5 Principle of Measurement

Fe (II) ion is complexed in an acidic (pH 2.90-3.50) medium with 1, 10-phenanthroline Chelate so as to form orange-red color. The color solution obeys the Beer's Lambert law.

6.2.6 Standards and Reagents

a) Preparation of 100mg/l of Iron (Fe) stock solution

Reagents

- Ammonium ferrous sulphate ($(NH_4)_2Fe(SO_4)_26H_2O$)
- Distilled water

Procedure

- Weigh 0.1755gm [$(NH_4)_2Fe(SO_4)_26H_2O$] .
- Transfer to 250 ml volumetric flask.
- Dilute to mark with distilled water.
- Transfer to an appropriately labeled bottle container with label.

b) Preparaton of 10 mg/l of Fe Solution and working standards

Reagents

- 100 mg/l Iron stock
- Distilled Water

Procedure

- Transfer 10 ml of 100-mg/l stock solutions in to 100 ml volumetric flask to prepare a 10 mg/l secondary standard.
- Dilute to mark with distilled water.
- Prepare standards of 0.05 ,0.1---0.3 ppm using formula $C_1V_1=C_2V_2$ by making dilution from 10 ppm Iron standard solution.
- Transfer to an appropriately labeled bottle container with label.

c) Preparation of phenonthroline solution for Iron (Fe)

Reagents

- 1,10-PHENANTHROLNE MONOHYDRATE solution
- Conc. Hydrochloric acid HCl

Procedure

- Weigh 100 mg $C_{12}H_8N_2.H_2O$.
- Transfer to 100 ml volumetric flask.
- Dissolve in 100 ml distilled water by making volume up to the mark.
- Add 2 drops of Conc. HCl (iron free).
- Transfer to an appropriately labeled bottle container with label.

d) Preparation of Ammonium acetate buffer solution for Iron (Fe)

Reagents

- Ammonium Acetate
- Conc. Glacial Acetic Acid
- Distilled Water

Procedure

- Weigh and dissolve 71.428g $NH_4C_2H_3O_2$ in 43 ml distilled water in 250 ml volumetric flask.
- Add 200 ml conc. Glacial Acetic Acid to it.
- Transfer to an appropriately labeled bottle container with label.

6.2.7 Calibration of Spectrophotometer for Iron

- Prepare iron standards from 0.05 to 0.5 ppm (0.05, 0.1, 0.15, 0.2, 0.25, 0.30, 0.35, 0.40, 0.45, and 0.5) from 1 ppm secondary standard.
- Now set the spectrophotometer according to equipment instructions by applying wavelength and standard correction.

- Take distilled water as blank and consider all standards as samples.
- Take 5 ml sample + 1 ml buffer +0.2 ml phenanthroline and take absorbance after 15 minutes at wavelength of 510 nm by constructing calibration curve.

6.2.8 Testing

- Take 10 ml deionized water in 50 ml cuvet, add 2 ml of Ammonium acetate buffer and 0.4 ml of 1,10-phenanthroline in it.
- Stir it vigorously and let it stand for 15 minutes (reaction time).
- Run the prepared blank solution on the spectrophotometer at the already prepared calibration curve and make special correction or blank correction.
- Then take 10 ml sample to be analyzed and treat it as done with blank.
- Number of replicates to be analyzed are “Three” for each sample.

6.2.9 Quality Control

- Method blank is analyzed after every 10 samples.
- Laboratory Control samples of 0.2 ppm analyzed with every batch of 10 samples.
- NWQL check repeatability by reanalyzing a pre-analyzed sample once a day.
- To ensure the accuracy and precision of method spiking with known standard solution done once in month.
- Daily quality control data is recorded in QC Data sheet-I.
- Results of laboratory control sample are regularly displayed in the form of control charts.

6.2.10 Expression of Results

The Iron is measured in mg/l.

6.2.11 Interpretation of Results

Compare the results with water quality standards or guidelines to predict the safe or unsafe level.

6.2.12 Precautions

- First check the reliability of the already saved curve by running number of known standards.
- The pH of the Ammonium acetate buffer solution must be in range (2.90-3.50).

6.2.13 Planned Demonstration

Testing of Iron in Chemical Lab III (Reagents preparation, glassware washing, standards preparation, testing, quality control, calculations and reporting, unit’s conversion).

6.2.14 Learning Outcomes

On successful completion of this course students will be able to:

1. Explain the importance of iron in water.
2. Identify the basic sources and impacts of high concentrations of Iron in water.
3. Prepare the reagents and working standards to construct the calibration curve.
4. Illustrate the procedure to calibrate the instrument for the determination of Iron.
5. Perform the testing of Iron for water samples.
6. Discuss water quality data, safe and unsafe levels.

6.2.15 Class Quiz

6.2.16 Group Exercise Water Quality Assessment

6.3 Alternate Method for Determination of Iron (II)

6.3.1 Definition

The purpose of this method is to characterize the quality of potable water in context to iron estimation in water.

6.3.2 Testing Instrument DR/2800 Colorimeter, HACH

6.3.3 Analytical Parametrs

- Fluoride
- Iron

6.3.4 Major Components

- Protective cover
- Source Lamp Tungsten
- square matched glass sample cells
- Cell adapters

6.3.5 Principle of Measurement

Fe (II) ion is complexed in an acidic (pH 2.90-3.50) medium with 1, 10-phenanthroline Chelate so as to form orange-red color. The color solution obeys the Beer's Lambert law. 1,10-Phenanthroline, contained in Ferrous Iron Reagent Powder, reacts with Fe^{2+} to form a characteristic orange-colored complex. The intensity of color development is directly proportional to the amount of Fe^{2+} in the sample.

6.3.6 Standards and Reagents

- FerroVer iron reagent
- HCL
- Distilled Water

6.3.7 Calibration of Colorimeter 2800 for Iron

- Turn on the power switch.
- Keep the lid covered. System will start the self check for lamp test, filter adjustment, wavelength adjustment and voltage test.
- Select the stored program for Iron.
- Press the start button.
- Prepare the working standard.
- Apply the blank sample in 25 ml curette give three minutes reaction time by pressing timer. After the timer beeps press zero then press read.
- Take 10 ml of standard solution maintain the ph by HCL then add the FrroVer pillow give three minutes reaction time then read the standard concentration after three minutes.

6.3.8 Testing

- Make blank correction, then take 10ml sample and treat it as described above and measure concentration.
- Number of replicates to be analyzed are “Three” for each sample.

6.3.9 Quality Control

- Method blank is analyzed after every 10 samples.
- Laboratory Control samples analyzed with every batch of 10 samples.
- NWQL check repeatability by reanalyzing a pre-analyzed sample once a day
- To ensure the accuracy and precision of method spiking with known standard solution done once in month.
- Daily quality control data is recorded in QC Data sheet-I.
- Results of laboratory control sample are regularly displayed in the form of control charts.

6.3.10 Expression of Results

The Iron is measured in mg/l.

6.3.11 Interpretation of Results

Compare the results with water quality standards or guidelines to predict the safe an unsafe level.

6.3.12 Precautions

- Check the calibration of instrument before and after use.
- Use samples and standards at the same temperature or at least within 2 °C difference.

6.3.13 Planned Demonstration

Testing of Iron in Chemical Lab III (Reagents preparation, glassware washing, standards preparation, testing, quality control, calculations and reporting, unit's conversion).

6.3.14 Learning Outcomes

On successful completion of this course students will be able to:

1. Explain the importance of iron in water.
2. Identify the basic sources and impacts of high concentrations of Iron in water.
3. Prepare the reagents and working standards.
4. Illustrate the procedure to calibrate the instrument for the determination of Iron.
5. Perform the testing of Iron for water samples.
6. Discuss water quality data, safe and unsafe levels.

6.3.15 Class Quiz

6.3.16 Group Exercise

Water Quality Assessment