

great care. The pH meter should be calibrated by checking against a standard buffer of known pH value. For quick calibration it is safer to use buffer tablets of known pH value for making standard buffer solution which is stored in refrigerator when in use. The electrodes must be washed thoroughly with distilled water, the solution of unknown pH placed in a glass beaker, the glass and calomel electrodes immersed in it and the pH on the meter scale read. Four general precautions should be taken.

1. The pH meter must be frequently checked against a standard buffer of known pH for scale setting.
2. The electrodes should be kept immersed in distilled water even when not in use.
3. The temperature of the solution under test must be recorded since temperature influences pH determination.
4. When lowering the electrodes inside the beaker, care should be taken not to touch the bottom or side walls of the beaker.

Whenever a solution in the beakers is changed for pH testing the electrodes must be washed with distilled water.

2. ANALYSIS OF WATER pH

Object :

To determine the pH of given water sample.

Apparatus and Glassware :

pH meter (any make), test tubes, graduated pipettes (1, 5 and 10 ml), reagent bottles, test tube stand, beakers, etc.

Principle :

The term acid/base balance has long been used in relation to the constancy of the hydrogen ion concentration in the body fluids. The pH may be expressed in terms of acidity which is simply the negative logarithm of the hydrogen ion concentration. Thus $\text{pH} = -\log (\text{H}^+)$. For example, pH of water is 0.0000001N, i.e., it contains 10^{-7} g equivalents of hydrogen per litre. The pH is stated to be 7.0 or $-\log (10^{-7})$ and represents neutrality. A pH lower than 7 indicates that hydrogen ion concentration is greater than that of water and the solution is an acid. A pH higher than 7 indicates a base. The usual range of pH scale is 0 to 14. Three methods can be employed to determine the pH.

A. pH Paper Strip Method :

Procedure :

1. Take given water sample.
2. Immerse a pH paper strip in the water sample.
3. Note the colour changes, match the colour and find out the pH with the help of the colour chart printed on the pH paper strip packet.

B. Universal Indicator Method :

Procedure :

1. Take 10 ml of given water sample in a clean and dried test tube.
2. Add 0.2 ml universal indicator solution and mix.
3. Match the colour of the solution of the test tube with that of the chart printed on the label of the bottle or follow the instructions of the manufacturer printed on the label of the bottle.

Result :

The matched colour will be the pH of the given water sample.

C. pH Determination by pH Meter :

Procedure :

This method (electronic method) gives an accurate determination of the pH of any solution. Several makes of pH meters are available in the market. The instrument consists of two electrodes, one glass electrode and other calomel electrode. Glass electrodes are not absolute in their measurement and should be repeatedly standardised with solutions of known pH which are commercially available. For operation of the pH meter, see the manual supplied with the instrument.

Note : For detail of the procedure follow the procedure as mentioned in the "Analysis of soil pH".

Result :

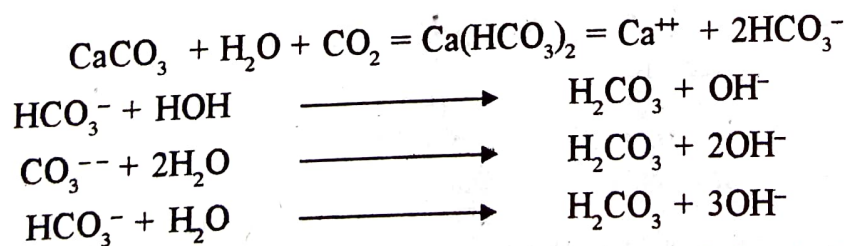
The reading in the pH meter will be the pH of given water sample.

3. ALKALINITY OF WATER

Object :

To estimate the alkalinity in given water sample.

Total alkalinity of water is the capability of neutralising or making ineffective completely the strong acids. Hydroxides (OH^-), carbonates (CO_3^{--}) and bicarbonates (HCO_3^-) contribute alkalinity to a liquid. CO_3^{--} and HCO_3^- produces alkalinity by combining with water in the following manner :



Alkalinity values provide guidance in applying proper doses of chemicals in water and wastewater treatment processes, particularly in coagulation, softening and operational control of anaerobic digestion.

Principle :

Alkalinity can be obtained by neutralizing OH^- , CO_3^{--} and HCO_3^- with standard H_2SO_4 . Titration to pH 8.3 or de-colourization of phenolphthalein indicator will show complete neutralization of OH^- and $\frac{1}{2}$ of CO_3^{--} , while to pH 4.4 or sharp change from yellow to pink of methyl orange indicator will indicate total alkalinity, *i.e.*, OH^- , CO_3^{--} and HCO_3^- .

Interference : As indicated in acidity estimation, Borate, Silicates and Phosphates also produce alkalinity.

Apparatus and Glassware :

Burette, stand, measuring cylinder (100 ml), beaker, titration flask, reagent bottles etc.

Reagents :

1. **Standard 0.02N H_2SO_4 :** Prepare 0.1N H_2SO_4 by diluting 3.0 ml Conc. H_2SO_4 to 1000 ml. Standardize it against standard Na_2CO_3 (0.1N). Dilute appropriate volume of H_2SO_4 (approx. 0.1N) to 1000 ml to obtain standard 0.02N H_2SO_4 .

OR

Standard 0.1 N Hydrochloric acid (0.1 N HCl) : Prepare 1.0 N HCl by diluting 8.34 ml 12 N Conc. HCl (specific gravity 1.18) to 100 ml distilled water or 25 ml 12 N Conc. HCl to 300 ml distilled water. Dilute so prepared 100 ml of 1.0 N HCl to 1000 ml distilled water to prepare 0.1 N HCl.

2. **Phenolphthalein indicator :** Dissolve 5 g in 500 ml 95% ethyl alcohol. Add 500 ml distilled water. Add dropwise 0.02N NaOH till faint pink colour appears.
3. **Methyl orange indicator :** Dissolve 0.5 g and dilute to 1000 ml with CO_2 free distilled water.

Procedure :

1. Take 25 or 50 ml sample in a conical flask and add 2-3 drops of **phenolphthalein indicator**.
2. If pink colour develops titrate with 0.02N H_2SO_4 till it disappears or pH is 8.3. Note the volume of H_2SO_4 required.

3. Add 2-3 drops **methyl orange** to the same flask, and continue titration till pH comes down to 4.5–4.4 or orange colour changes to pink.

Note the Volume of H_2SO_4 added.

4. In case pink colour does not appear after addition of **phenolphthalein** continue as in procedure point No. 3 above.

Observation :

Prepare the observation table as mentioned below :

S. No.	Water sample taken (ml)	Burette Reading (ml)					
		with phenolphthalein			with methyl orange		
		Initial	Final	Total (A)	Initial	Final	Total (B)
1.							
2.							
3.							
4.							

Calculation :

1. Calculate **Total (T), phenolphthalein (P) and methyl orange (MO) alkalinity** as follows and express result as mg/l as CaCO_3 :

P alkalinity, mg/l as $\text{CaCO}_3 = A \times 1000/\text{ml sample}$

MO alkalinity, mg/l as $\text{CaCO}_3 = B \times 1000/\text{ml sample}$

T alkalinity, mg/l as $\text{CaCO}_3 = (A + B) \times 1000/\text{ml sample}$

In case H_2SO_4 is not 0.02N apply the following formula :

Alkalinity mg/l as $\text{CaCO}_3 = \frac{A \times N \times 50000}{\text{ml sample}}$

where N = Normality of H_2SO_4 used.

Calculate OH^- , CO_3^{--} and HCO_3^- forms from the values of P and T alkalinity as shown below :

Values of P & T	OH^- Alkalinity as $\text{Ca}(\text{OH})_2$	CO_3^{--} Alkalinity as CaCO_3	HCO_3^- Alkalinity as $\text{Ca}(\text{HCO}_3)_2$
$P = 0$	0	0	T
$P < \frac{1}{2} T$	0	2P	$T - 2P$
$P = \frac{1}{2} T$	0	2P	0
$P > \frac{1}{2} T$	$2P - T$	$2(T - P)$	0
$P = T$	T	0	0

Here P means = Phenolphthalein Alkalinity

and T denotes = Total Alkalinity.

After estimation with Phenolphthalein indicator, in the sample of the same solution, methyl orange is added and still the colour remains orange, it indicates **Phenolphthalein** alkalinity is the **Total alkalinity**. It does not mean the Total Alkalinity is zero (0).

Result :

The alkalinity of given water sample = mg/l

4. ACIDITY OF WATER

Object :

To estimate the acidity of given water sample.

Acidity of a liquid is its capacity to donate H^+ ions. Since most of the natural waters and sewages are buffered by carbon dioxide-bicarbonate system, the acidity present due to free CO_2 has no significance from public health point of view. Waters containing mineral acidity (due to H_2SO_4 and HCl) are unacceptable. Further, acid waters pose problem of corrosion and interfere in water softening and water treatment processes.

Principle :

The mineral acids present and contributing mineral acidity can be calculated by titrating or neutralizing samples to pH 4.3. The CO_2 and bicarbonates (carbonic acid) present in the sample can be neutralized completely by continuing the titration to pH 8.3.

Interference : Colour, turbidity, iron, aluminium and residual chlorine and prime sources of interference. Colour and turbidity can be avoided using potentiometric titrations. Residual chlorine can be removed by adding thiosulphate.

Apparatus and Glassware : Burette, stand, measuring cylinder (100 ml), beaker, titration flask, reagent bottles etc.

Reagents :

1. **Standard sodium hydroxide 0.02N :** Dissolve 0.8 g NaOH and dilute to 1000 ml using CO_2 free distilled water. Store in air-tight, rubber stoppered pyrex/corning glass bottle to protect from atmospheric CO_2 . Standardize against 0.02N potassium biphthalate.
2. **Phenolphthalein indicator :** Dissolve 5 g in 500 ml 95% ethyl alcohol. Add 500 ml distilled water. Add dropwise 0.02 NaOH till faint pink colour appears.
3. **Methyl orange indicator :** Dissolve 0.5 g and dilute to 1000 ml CO_2 free distilled water.

Procedure :

1. Measure suitable volume of sample (50 or 100 ml) in 250 ml conical flask or beaker depending upon the method to be followed.
2. Add 2 drops of **methyl orange** and titrate with standard 0.02 N NaOH till colour changes to faint orange characteristic of pH 4.4-4.3.
3. Note down the volume of NaOH used.

4. Add 2-3 drops **phenolphthalein** indicator and continue titration with NaOH till faint pink colour appears indicating pH 8.3.
5. Note down the volume of additional NaOH used.

Observation :

Prepare the observation table as metioned below :

S. No.	Water sample taken (ml)	Burette Reading (ml)					
		with methyl orange			with phenolphthalein		
		Initial	Final	Total (A)	Initial	Final	Total (B)
1.							
2.							
3.							

Calculation :

- (i) Each ml of 0.02 NaOH is = 1 mg CaCO_3

Therefore, acidity of mineral or due to CO_2 as mg/l CaCO_3 = the used volume (ml) of 0.02 NaOH \times 1000 ml sample.

In case of normality of NaOH is other than 0.02, calculate as follows :

- (ii) Acidity of mineral or due to CO_2 as mg/l CaCO_3 .

$$= \frac{A \text{ or } B \times N \times 50000}{\text{ml sample}}$$

where;

A = ml NaOH used for sample to raise pH up to 4.4-4.3

B = ml NaOH used for sample to raise pH for 4.4 to 8.3

N = normality of NaOH used.

Result :

The acidity of given water sample = mg/l

5. DISSOLVED OXYGEN (DO)

Object :

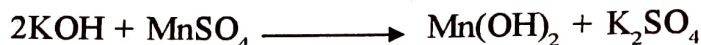
To estimate dissolved oxygen in given water sample.

Living organisms need oxygen to maintain their metabolic processes. Dissolved oxygen (DO) is also important in precipitation and dissolution of inorganic substances in water. The solubility of

oxygen in water depends upon its temperature. Analysis of DO is a key test in sanitary engineering practice. Therefore, it necessary to know DO levels to assess quality of raw water and to keep a check on stream pollution.

Principle :

In a dissolved state in water O_2 is not reactive in the molecular form. Therefore, to make it reactive, $Mn(OH)_2$ (Manganese hydroxide) is used. It is obtained from $MnSO_4$ in the presence of an alkali as follows :



This $Mn(OH)_2$ combines with O_2 and forms a brown precipitate (ppt) of Manganic oxide $MnO(OH)_2$, as follows :



Thus, ppt of manganic oxide reacts with H_2SO_4 and produces nascent oxygen (O). Since, nascent oxygen is highly reactive, it dissociates iodine from KI in the presence of H_2SO_4

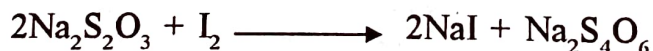


or



Iodine turns starch blue, with the help of which it can be estimated that equivalent quantity of starch turned blue will indicate the amount of iodine.

Iodine reacts with sodium thio-sulphate which does not give blue colour with starch. Therefore, when all the I_2 turns into NaI , then solution becomes colourless.



Note : Many times it has been observed that few ions, e.g., NO_2^{--} , SO_3^{--} which remain present in water, can free iodine from KI just like that of nascent oxygen (O). Their presence is always a hurdle in the estimation of real amount of dissolved O_2 . Therefore, to remove this problem, sodium azide (NaN_3) is added in solution before adding H_2SO_4 .



Apparatus and Glassware :

Burette, stand, measuring cylinder (100ml), beaker, titration flask, reagent bottles, etc.

Reagents :

- Manganous sulphate :** Dissolve 48 g tetrahydrate manganous sulphate ($MnSO_4 \cdot H_2O$) and dilute to 100 ml. Filter if necessary. This should not give colour with starch added to an acidified solution of potassium iodide (KI).

2. **Alkali iodide-azide reagent** : Dissolve 500 g NaOH and 150 g KI and dilute to 1000 ml. Add 10 g NaN_3 dissolved in 40 ml distilled water. This solution should not give colour with starch solution when diluted and acidified.
3. Conc H_2SO_4
4. **Starch indicator** : Prepare paste or solution of 0.5 g starch powder in distilled water. Pour this solution in 100 ml boiling water. Allow to boil for few minutes. Cool and then use. This can be stored by adding few drops of toluene or formaldehyde.
5. **Stock sodium thiosulphate, 0.1N** : Dissolve 24.82 g $\text{Na}_2\text{S}_2\text{O}_3 \cdot 5\text{H}_2\text{O}$ in boiled cooled distilled water and dilute to 1000 ml. Preserve by adding 5 ml chloroform per litre or 3 gm borax.
6. **Standard sodium thiosulphate, 0.025N** : Dilute 250 ml stock $\text{Na}_2\text{S}_2\text{O}_3$ solution to 1000 ml with freshly boiled and cooled distilled water. Preserve by adding 5 ml chloroform per litre. (This solution will have to be standardized for each set of titrations).

Procedure

1. Collect sample in a BOD bottle using DO samples.
2. Add 2 ml MnSO_4 solution followed by 2 ml of alkali iodide-azide solution. The tip of the pipette should be below the liquid level while adding these reagents. Stopper immediately.
3. Mix well by inverting the bottle 2-3 times and allow the precipitate to settle leaving 150 ml clear supernatant.
4. At this stage, add 1 ml Conc. H_2SO_4 . Mix well till precipitate goes into solution. Avoid any kind of air bubble in the bottle. By adding H_2SO_4 brown ppt. is dissolved and nascent O is formed which releases I_2 from KI. Due to this reaction solution appears yellow.
5. Take 203 ml of this solution in a conical flask and titrate against $\text{Na}_2\text{S}_2\text{O}_3$ using starch as an indicator. When 2 ml MnSO_4 followed by 2 ml alkali iodide-azide solution is added to the sample as in (step) above, 4.0 ml of original sample is lost. Thus, 203 ml taken for titration will correspond to 200 ml of original sample.

$$200 \times 300 / (300 - 4) = 203 \text{ ml}$$

Observation :

Record your reading in the following table :

S. No.	The volume of sample (A)	Burette reading			Mean of Thiosulphate volume (B)
		Initial	Final	Total	
1.					
2.					
3.					
4.					

$$\text{Dissolved O}_2 \text{ (mg/l)} = \frac{\text{Volume of Thiosulphate used (ml) B} \times \text{Normality} \times 1000 \times 8}{\text{Volume of sample measured (A)}}$$

At NTP, the dissolved O₂ content (ml/l) is found by following formula :

$$= \frac{0.2 \times 1000 \times 0.698}{200 \text{ ml (i.e., sample volume)}} \text{ is the present expression.}$$

(in this manner mg/l estimation can be converted into ml/l).

Calculation :

For example :

1 ml of 0.025N Na₂S₂O₃ = 0.2 mg of O₂

Since the sample volume is 200 ml, $\frac{(0.2 \times 1000)}{200}$

1 ml of thiosulphate = DO mg/l. Each mole of gas at NTP occupies 22.4 l. Since the mole of O₂ weighs 16, multiply this value (DO mg/l) by 0.698 to give ml of O₂ present in the sample at 0°C and 760 mm (16/22.4 = 0.698).

Result :

The dissolved oxygen of given water sample = mg/l

Precautions :

1. Thiosulphate solution should be prepared fresh only. Its standard solution can, however, be used for 2 weeks.
2. For taking and using the H₂SO₄, never use sucking by mouth. Always use rubber bulb pipette.
3. Avoid air bubbles at various stages.

6. FREE CARBON DIOXIDE (CO₂)

Object :

To estimate free carbon dioxide in a given water sample.

Principle :

Free CO₂ can be determined by titrating the sample using a strong alkali (e.g., carbonate free NaOH) to pH 8.3. At this pH, the entire free CO₂ is converted into bicarbonates.

Apparatus and Glassware :

Burette, stand, measuring cylinder (100 ml), beaker, titration flask, reagent bottles, etc.

Reagents :

1. NaOH, 0.05N : Prepare 1.0N NaOH by dissolving 40 g of NaOH in CO₂ free distilled

water to make 1 litre of solution. Dilute 50 ml of 1.0N NaOH to 1 litre. Standardise it with H_2SO_4 , HCl or Oxalic acid.

2. **Phenolphthalein Indicator** : Dissolve 0.5 g of phenolphthalein in 50 ml of 95% ethanol and add 50 ml of distilled water. Add 0.05N CO_2 free NaOH solution dropwise until the solution turns faintly pink.

Procedure :

1. Take 100 ml of sample in a conical flask and add a few drops of phenolphthalein indicator. If the colour turns pink, free CO_2 is absent. If the sample remains colourless titrate it against 0.05N NaOH. At the end point a pink colour appears.

Observation :

Prepare the observation table as mentioned below :

S. No.	Water sample taken (ml)	Burette Reading (ml)			Mean (ml)
		Initial	Final	Total	
1.					
2.					
3.					

Calculation :

$$\text{Free CO}_2, \text{ mg/l} = \frac{(\text{ml} \times \text{N}) \text{ of NaOH} \times 1000 \times 44}{\text{ml sample}}$$

Result :

The free carbon dioxide of given water sample = mg/l

7. CHLORIDES (SALINITY)

Object :

To estimate chloride (salinity) in a given water sample.

Chlorides occur widely in water and waste waters and are usually associated with Na ion. Although, chlorides are not harmful, concentrations beyond 250 mg/l impart a peculiar taste to water rendering it unacceptable from aesthetic point of view for drinking purpose. Presence of chlorides above the usual background concentration in a water source is also used as an indicator of pollution by domestic sewage.

Principle :

Chloride ion is determined by titration with standard AgNO_3 in which AgCl_2 precipitates out.



As per ICMR (1975) guidelines, chlorinity in drinking water should not be more than 1000 mg / l.

The end of titration with potassium chromate is indicated by formation of red silver chromate from excess AgNO_3 . Potassium chromate is used as an indicator in neutral to slightly alkaline solution.



Interference : Bromide, iodide, cyanide, sulphide, thiosulphate, sulphite, iron, phosphate are prime source of interference.

Apparatus and Glassware :

Burette, stand, measuring cylinder (100 ml), beaker, titration flask, reagent bottles, etc.

Reagents :

- Potassium chromate indicator (5%) :** Dissolve 50 g K_2CrO_4 in distilled water in 1000 ml. Add AgNO_3 till definite red precipitate is formed. Allow to stand for 12 hrs. Filter and dilute to 1000 ml.
- Silver nitrate, 0.0141N :** Dissolve 2.395 g AgNO_3 and dilute to 1000 ml. Standardize against NaCl, 0.0141N. 1 ml of 0.041N AgNO_3 = 0.5 mg NaCl.
Store this solution in coloured glass bottle wrapped with black paper.
- Sodium chloride 0.041N :** Dissolve 824.1 mg NaCl (dried at 140°C) and dilute to 1000 ml. = 0.5 mg NaCl.
- Special reagent to remove colour and turbidity :** Dissolve 125 g $\text{AlK}(\text{SO}_4)_2 \cdot 12\text{H}_2\text{O}$ or $\text{AlNH}_4(\text{SO}_4)_2 \cdot 12\text{H}_2\text{O}$ and dilute to 100 ml. Warm to 60°C and add 55 ml Conc. NH_4OH slowly. Allow to stand for 1 hr. Solution should be free from chloride.

Procedure :

- (1) Pretreatment : Take 100 ml sample and add 3 ml special reagent. Mix well and allow to settle. Filter the supernatant for titration purpose.
- (2) To the samples having sulphite neutralize the sample and add about 1ml H_2O_2 .
- (3) In case if sulphide or thiosulphate is present raise the pH of the sample to 8.3 or more, and H_2O_2 1 ml, and then again neutralise the sample.

(b) Titration :

1. Adjust the pH of sample between 7.0 and 8.0.
2. Take 50 ml well mixed sample adjusted to pH 7.0 - 8.0 and add 1.0 ml K_2CrO_4 .
3. Titrate with standard AgNO_3 solution till AgCrO_4 starts precipitating.
4. Standardize AgNO_3 against standard NaCl.
5. For better accuracy titrate distilled water (50 ml) in the same way to establish reagent blank.

Observation :

Prepare the observation table as mentioned below :

S. No.	Sample taken (ml)	Burette Reading (ml)			Mean (ml)
		Initial	Final	Total	
1.					
2.					
3.					

Calculation :

$$\text{Cl mg/l} = \frac{C \times N \times 35.450}{\text{ml sample}}$$

where C = ml AgNO₃ used

N = Normality of AgNO₃

Result :

The chlorides in given water sample = mg/l

8. QUALITATIVE ESTIMATION OF ZOO-PLANKTON IN GIVEN SAMPLE OF WATER

Object : To qualitatively estimate the zooplankton in given sample of water.

Principle :

Ponds and ditches containing organic matter which contain many micro-organisms. These may be observed in living condition.

Apparatus and Glassware :

Glass slides, cover slips, compound microscope, beaker, reagent bottles, glass dropper, etc.

Reagent :

4% Methyl cellulose solution :

Methyl cellulose

4 g

Distilled water

100 ml

Mix.

Procedure :

1. Collect some water from the bottom of a pond or a ditch containing rich and decaying organic matter in a beaker.
2. Put a drop of so collected water on a clean glass slide and a drop of 4% methyl cellulose solution. Adding methyl cellulose in the drop will slows down the movement of micro-organisms present in the water sample.
3. Cover the water drop with the cover slip.