### Aim - Estimation of Peroxide Value

Rancidity is brought about by the action of air (oxidative rancidity), hydrolysis of the lipid components or by microorganisms (ketonic rancidity) in oil. In oxidative rancidity oxygen is taken up by the fat with the formation of peroxides. Oxidative rancidity occurs when highly unsaturated fatty acids are exposed to oxygen due to oxidative hydroperoxides are formed. The degree of peroxide formation and the time taken for the development of rancidity differ among oils. Measurement of peroxide value indicates the initial rancidity status as it is the intermediate product for further degradation to carbonyl compounds such as aldehydes.

**Principle:** *The peroxides present are determined by titration against thiosulphate in the presence of KI. Starch is used as indicator.* The peroxide value of an oil or fat is the amount of peroxides (generated during oxidation) present in the fat and expressed as milli-equivalents of peroxide oxygen per kilogram of fat.

### Materials:

- Solvent: Mix two volumes of glacial acetic acid and 1 volume of chloroform.
- Saturated potassium iodide solution: KI is dissolved in distilled water in 4:3 ratio and stored in brown bottle.
- 0.01N sodium thiosulphate solution. Prepare N/10 solution and dilute on the day of use.
- 1% starch solution (to be prepared fresh).
- Anhydrous sodium sulphate (should be heated at 180°C for 3 hours and collected in desiccator before use).

## Method:

### Preparation of chloroform extract for fish muscle:

- Take 10g of sample with 15g anhydrous sodium sulphate and grind well using a mortar and pestle.
- Transfer the grounded mixture in to a 250 ml iodine flask.
- Then, add about 80-100 ml of chloroform to the flask and shake for 2-3 min.
- Keep in dark overnight.
- Filter the filtrate by Whatman No.1 filter paper taking anhydrous sodium sulphate in it.

• This chloroform extract can be use to estimate peroxide value and free fatty acid value.

## **Estimation:**

- Take 1-2g of fish oil in a 250 ml iodine flask.
- To it add 25 ml of solvent to dissolve the fat with thorough shaking.
- In case of fish muscle, take 5-10 ml of chloroform extract in a 250ml iodine flask and add 25ml of solvent to it.
- Then add 1 ml of saturated potassium iodide (KI) solution (or about 1g KI salt).
- Shake the content vigorously for a minute and allow standing in dark for 30 min.
- After 30 minutes add about 35 ml of distilled water by washing the stopper and sides of the flask.
- Titrate the contents of the flask against 0.01N sodium thiosulphate solution using starch as an indicator
- Vigorous shaking is required to liberate all iodine from chloroform layer till first complete disappearance of blue colour.
- Run a blank simultaneously with solvent only.

# **Calculation:**

Peroxide value (milli-equivalent peroxide O<sub>2</sub> per 1000g fat) =

Where,

- S = Vol. of Sodium Thiosulphate (sample-blank)
- N = Normality of Sodium Thiosulphate
- W = Weight of sample