: Karl Fischer Titration:

BY:Ranjeet Ranjan

Introduction: Aquametry can be defined as quantitative determination of water in Pharmaceuticals.

The Karl Fischer Titration (KFT) technique was introduced in 1935, for the determination of small amounts of water in a variety of organic and inorganic solid and liquid samples. The fundamental principle behind it is based on the Bunsen Reaction between iodine and sulfur dioxide in an aqueous medium. Karl Fischer discovered that this reaction could be modified for the determination of water in a non-aqueous system containing an excess of sulfur dioxide. He used a primary alcohol (methanol) as the solvent, and a base (pyridine) as the buffering agent.

Background: Water, in small amount, is generally controlled by a loss on drying under specified conditions. When present in appreciable amount, the water content may often be readily determined by titration with KARL FISSHER REAGENT, which contains iodine, sulphur dioxide, anhydrous methanol and anhydrous pyridine.

The reaction of Karl Fischer Titration is a three-stage process. The first stage involves the reaction of sulphur dioxide with water and iodine which reduces iodine (1), in second stage SO_3 forms a complex with pyridine (2)

$SO_2 + I_2 + H_2O \rightleftharpoons SO_3 + 2HI$ -------------------------- (1)

$C_5H_5N + SO_3 \longrightarrow C_5H_5N^+ . SO_2.O$------------------------- (2)

The third stage involves reaction of pyridine complex with methanol to form stable complex (3).

$C_5H_5N^+ . SO_2.O^- + CH_3OH \longrightarrow C_5H_5N^+HCH_3O.SO_2.O$--------------------- (3)

The each molecule of iodine is equivalent to one molecule of water.

The reactive alcohol is typically methanol or 2-(2-Ethoxyethoxy) ethanol, also known as diethylene glycol monoethyl ether (DEGEE), or another suitable alcohol. Classic Karl Fisher reagents contained pyridine, a noxious carcinogen, as the base. The reagents most frequently used today are pyridine-free and contain imidazole or primary amines instead.

There are two important requirements if the above reactions are to be stoichiometric. Firstly, the alcohol used must completely etherify the sulphur dioxide and secondly the base must be of suitable strength to completely neutralize the acids produced during the reaction.

Preparation of Karl Fischer Reagent:

Method – Into a glass-stopper flask (about 750 ml capacity) place anhydrous methanol (400 ml, containing not more than 0.03 % water) and pure dry pyridine 80 mg. Immerse the flask in a freezing bath and slowly pass sulphur dioxide into a cold solution, with continuous agitation, until the increase in the weight is 20 gm. Care must be taken all time to prevent access of moisture. Finally add iodine 45 gm, shake to dissolve and allow the mixture to stand for 24 h before use. The reagent prepared in this manner will have water equivalent of 3.5 mg/ml approximately.

Standardisation: - Karl Fischer reagent is not stable and must be rigorously protected from both light and moisture. It must be standardized immediately before use by titration against a known amount of water, either as a standard water-methanol reagent or, preferably, as solid crystalline sodium tartrate dihydrate. Although the latter is almost insoluble in methanol, it yields its water of hydration to the solvent, and its precise water content can be determined by drying at 150^0 to constant weight.

How does it work: Water and iodine are consumed in a 1:1 ratio in the above reaction. When all of the water present is consumed, the presence of excess iodine is detected by indicator electrode that signals the end-point of the titration. The amount of water present in the sample is calculated, based on the concentration of iodine in the Karl Fisher titrating reagent (i.e., titer) and the amount of Karl Fisher Reagent consumed in the titration.

Role of pH: The rate of the reaction depends on the pH value of the solvent or working medium. The titration proceeds normally when pH is between 5 to 8. However, when the pH is lower than 5, the titration speed is very slow. On the other hand, when pH is higher than 8, titration rate is fast, only due to an interfering side reaction, which produces water, resulting in a vanishing endpoint. Thus, the optimal pH range for the Karl Fischer reaction is from 5 to 8, and highly acidic or basic samples need to be buffered to bring the overall pH into that range.

Karl Fischer Instrument: The Karl Fischer instrument used is the Mitsubishi Coulometric moisture meter Model CA-100.

Titration Method: Electrolytic oxidation is carried out in the analyte, which consists mainly of iodide, sulphur dioxide, bases, and solvents such as alcohols. Electrolytic oxidation proceeds when a sample is added to the analyte. The Karl Fischer reaction occurs to produce iodine.

$$2I^- + 2e \longrightarrow I_2$$

Iodine is produced in proportion to the quantity of electricity so the water content can be estimated based on the coulombs required for electrolytic oxidation. A check solution containing a known amount of water is titrated at the beginning and the end of experiments to ensure that the KF instrument is functioning correctly.

Types of Karl Fischer Titrations:

1) **Volumetric KFT:** In volumetric Karl Fischer, iodine is added mechanically to a solvent containing the sample by the titrator's burette during the titration. Water is quantified on the basis of the volume of Karl Fischer reagent consumed. Volumetric analysis is best suited for determination of water content in the range of 100 ppm to 100%.

There are two main types of volumetric KFT reagent systems:

a) <u>In one-component volumetric</u> KF, the titrating reagent (also known as a CombiTitrant, or a Composite) contains all of the chemicals needed for the Karl Fischer Reaction, namely iodine, sulfur dioxide, and the base, dissolved in a suitable alcohol. Methanol is typically used as the working medium in the titration cell. One-component volumetric reagents are easier to handle, and are usually less expensive than two-component reagents.

b) <u>In two-component volumetric</u> KF, the titrating agent (usually known as the Titrant) contains only iodine and methanol, while the Solvent containing the other Karl Fischer Reaction components is used as the working medium in the titration cell. Two-component reagents have better long-term stability and faster titration times than one-component reagents, but are usually more costly, and have lower solvent capacity.

1) **Coulometric KFT:** In coulometric Karl Fischer, iodine is generated electrochemically *in situ* during the titration. Water is quantified on the basis of the total charge passed (Q), as measured by current (amperes) and time (seconds), according to the following relationship:

$$Q = 1\ \text{C (Coulomb)} = 1\ \text{A} \times 1\ \text{s} \quad (\text{where } 1\ \text{mg } H_2O = 10.72\ \text{C})$$

Coulometry is best suited for determination of water content in the range of 1 ppm to 5%. There are two main types of coulometric KFT reagent systems:

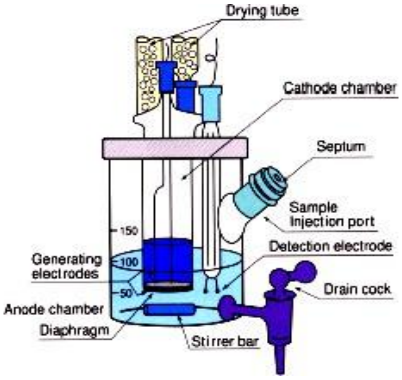
a) In conventional, or fritted-cell, coulometric KF, a diaphragm – or frit – separates the anode from the cathode that forms the electrolytic cell known as the generator electrode. The purpose of the frit is to prevent the iodine generated at the anode from being reduced back to iodide at the cathode instead of reacting with water.

b) In fritless-cell coulometric KF, an innovative cell design is used that through a combination of factors, but without a frit, makes it nearly impossible for iodine to reach the cathode and get reduced to iodide instead of reacting with water.

The advantages of the fritless cell include:

- Uses only one reagent
- Lower reagent cost
- Titration cell much easier to clean
- Reduced downtime
- Lower maintenance cost
- Long-term drift (background) value more stable
- Can use reagent longer without refilling
- Refilling of electrolyte suitable for automation
- Reduced downtime
- Increased lab safety

Titration Cell for Coulometric Titration



Working of volumetric titrator: The volumetric titrator performs the following three key functions:

- It dispenses KF titrating reagent containing iodine into the cell using the burette
- It detects the endpoint of the titration using the double platinum pin indicator electrode.
- It calculates the end result based on the volume of KF reagent dispensed using the On-board microprocessor.

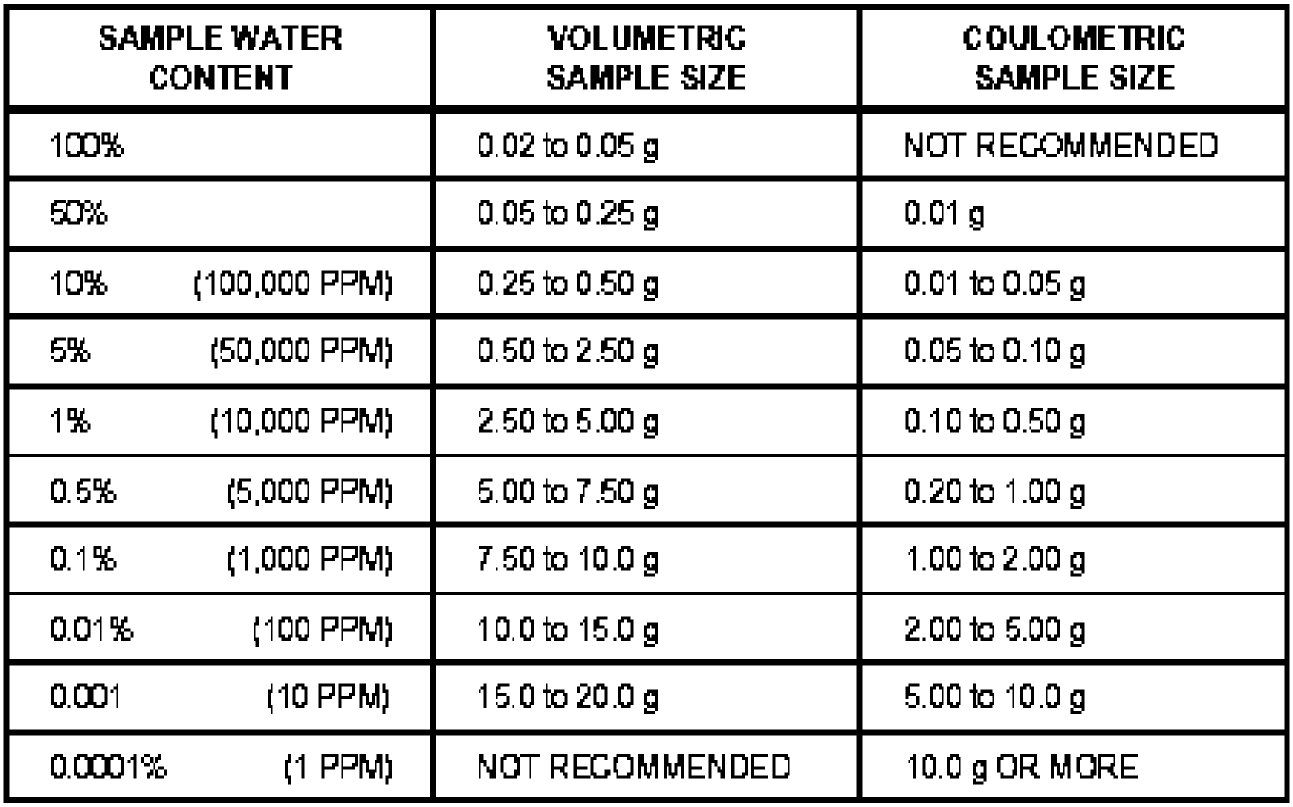
Working of Coulometric Titrator: The titrator performs the following three key functions:

1) It generates iodine at the anode of the titration cell, instead of dispensing KF reagent as in volumetric titration

2) It detects the endpoint of the titration using the double platinum pin indicator electrode

3) It calculates the end result based on the total charge passed (Q), in Coulombs, using the on- board microprocessor

4) **Sample size:** The amount of sample used depends on the anticipated water content and the desired degree of accuracy. Refer to the following convenient reference table:



SAMPLE WATER CONTENT		VOLUMETRIC SAMPLE SIZE	COULOMETRIC SAMPLE SIZE
100%		0.02 to 0.05 g	NOT RECOMMENDED
50%		0.05 to 0.25 g	0.01 g
10%	(100,000 PPM)	0.25 to 0.50 g	0.01 to 0.05 g
5%	(50,000 PPM)	0.50 to 2.50 g	0.05 to 0.10 g
1%	(10,000 PPM)	2.50 to 5.00 g	0.10 to 0.50 g
0.5%	(5,000 PPM)	5.00 to 7.50 g	0.20 to 1.00 g
0.1%	(1,000 PPM)	7.50 to 10.0 g	1.00 to 2.00 g
0.01%	(100 PPM)	10.0 to 15.0 g	2.00 to 5.00 g
0.001	(10 PPM)	15.0 to 20.0 g	5.00 to 10.0 g
0.0001%	(1 PPM)	NOT RECOMMENDED	10.0 g OR MORE

Limitation of Karl Fischer Titration: The Karl Fischer titration has a number of serious limitation due to possible interference to erroneous results, namely:

Oxidizing agents, for instance: chromates, Cu (II), Fe (III), peroxides, salts and higher oxides.
Example: $MnO_2 + 4\ C_5H_5NH^+ + 2I^- \rightarrow Mn^{++} + 4C_5H_5N + I_2 + H_2O$

Reducing agent, such as: Sn (II) salts, sulphides and $S_2O_3{}^{-2}$.

Compound that have a tendency to form water with the ingredients of the KFR,
for instances: Basic oxides – eg. ZnO
Salts of weak oxy-acids –eg $NaHCO_3$

Uses of Karl Fischer Method: The method is used to control the water content of –

- Betamethasone sodium phosphate
- Cloxacilline sodium
- Cyclophosphamide
- Cyclophosphamide injections
- Prednisolone sodium phosphate
- Procaine penicillin
- Fortified procaine penicillin injection