Unit - 1st

* Pharmacopoeia :-

→ The term Pharmacopoeia comes from the Greek word "pharmakon" meaning drug and "poieo" meaning make, and the combination means "any formula or standards required to make a drug.

→ It is a book containing collection of monographs and published by an authorized body like government or Pharmaceutical society.

→ Pharmacopoeia is the official book of standards for drugs prepared by any country or regulatory body to specify the standards of identity, purity and strength for the drugs imported, manufactured or distributed throughout the country or a specific region.

- A monograph is a collection of detailed information on a particular drug, its dosage forms and methods of analysis.
- A Monograph contains:-

(1) chemical name
(2) formula
(3) solubility
(4) Identification
(5) pH
(6) Assay
(7) Specific optical rotation
(8) Loss on drying.
(9) sulphated ash
(10) Dose.

History of Pharmacopoeia :-

→ Each country has legislation on pharmaceutical preparations which sets standards and required quality for medicament, raw materials and preparations employed in the manufacture of drugs.

→ These regulation are presented in separate articles.

→ General and specific matters relating to individual drugs are published in the form of a book called a Pharmacopoeia.

- On 15th December 1820, the first united state pharmacopoeia (U.S.P) was released.
- In 1864, the first British Pharmacopoeia (B.P) was Published with inclusion of monographs on benzoic acid, Gallic acid, tartaric acid, tannic acid, camphor, lactose, sucrose and seven alkaloids along with their salts.

Indian Pharmacopoeia :-

→ The government of India constituted a permanent Indian Pharmacopoeia committee in 1948 under the chairmanship of R. N. Chopra.

→ For the preparation of the Indian Pharmacopoeia and established a central Indian pharmacopoeia laboratory at Ghaziabad uttar pradesh to keep it up to date.

→ The first eddition of the I.P was published in the year 1955 under the chairmanship of Dr. B.N. Ghosh.

→ The second edition of I.P in 1966 with some modification under the chairmanship of Dr. B. Mukerji.

→ The third edition of the I.P was published in 1985 under the chairmanship of Dr. Nityanand.

- In this pharmacopoeia inclusion of traditional system of drugs was made.

→ The fourth edition was published eleven year later, followed by the addendums published first in 2000 and the in 2002 and 2005. In addition supplement 2000 for vitrinary products were also released.

- **Different types of Pharmacopoeia:-**

(1) United state pharmacopoeia (USP) (1820)
(2) Indian pharmacopoeia (IP). (1948)
(3) German Pharmacopoeia (1872).
(4) British pharmacopoeia. (BP) (1864)
(5) Mexican Pharmacopoeia (1946)
(6) French Pharmacopoeia.
(7) Japanese Pharmacopoeia. (1886)
(8) European Pharmacopoeia. (1964)

Sources of Impurities :-

→ The purpose of drug substances or drug formulations (pharmaceuticals) is primarly for the well-being of humans. They cure patients of diseases, disorders or deficiencies.

→ They cure due to their potency and therapeutic efficacy.

→ Today a large number of drugs, chemicals and other substances are used in formulation. It is however, pertinent that pharmaceutical chemicals and formulation must maintain a very high degree of purity.

→ A compound is said to be impure if it has foreign matter / impurities. These impurities affect its potency.

Effect of Impurities on Pharmaceuticals :-

(1) Some impurities if present beyond certain tolerance limits can cause unwanted side effects that can lead to unpleasant reactions.
e.g → Heavy metals like Pb, Fe and Ag salts.

(2) Some impurities which are otherwise harmless in nature and without any therapeutic effects, if present in considerable proportions dilute the active strength or potency of the drug substance.
e.g → Na, K, Cl, SO_4, CO_3 salts.

(3) Some impurities may be able to catalyse the degradation, thereby shortening the shelf life of the drug substance.

④ Some impurities by their chemical nature can interact with the drug substance to affect its purity and potency. such impurities are said to be incompatible with the drug substance.

⑤ Some impurities by virtue of other unstable nature like hygroscopic nature, oxidisable nature etc. can bring about change in appearance, taste, odour, stability etc of the drug substance causing technical difficulties in its use as well as formulation.

- Impurities may entir or are formed in a drug substance during any of the following three stages.

① During manufacturing
② During Purification and processing.
③ During storage.

① During manufacturing :-

ⓐ Raw materials employed :-

Impurities present in raw materials may be carried through the manufacturing process to contaminate the final product.

→ Impurities such as As, Pb, heavy metals, chlorides associated with Na compound.

ⓑ Reagent used in manufacturing process

→ If reagent used in the manufacturing process contain some impurities these may find entry into the final product.

→ For e.g- H_2SO_4 is used in many chemical processes. This acid often has lead present in it. Anions like Cl^- and SO_4^{-2} are common impurities in many substances because of the use of hydrochloric acid and H_2SO_4 respectively in processing.

© Solvents used in the manufacturing process :-

→ If proper quality / purity of solvents is not assured, they may add to the impurities. Solvents like toluene, n-butanol contain water as an azeotrope.

→ Alcoholic solvents also may be contaminated with water and ethyl acetate can contain acetic in small amounts. Thus quality of solvents needs to be assured and controlled.

(d) Reaction vessels :-

→ Some solvents and reagents employed in the process may react with the metals of the reaction vessels, leading to their corrosion and passing traces of metal impurities into the solution, contaminating the final product.

(e) Intermediates :-

→ Sometimes, an intermediate substance produced during the manufacturing process may contaminate the final product.

$$NaBrO_3 + 3C \rightarrow NaBr + 3CO$$

→ If sodium bromate is not completely converted to the sodium bromide then it is likely to be present as an impurity.

(f) Atmospheric contamination :-

→ Atmosphere may contain dust (aluminium oxide, sulphur, silica, soot etc) and some gases like carbon dioxide, sulphur dioxide, arsine and hydrogen sulphide.

→ These may contaminate the final product during the manufacturing process.

→ e.g → NaOH readily absorbs atmospheric carbon dioxide when exposed to atmosphere.

(g) Defects in manufacturing process :-

→ Defect like imperfect mixing non-adherence to optimum reactions conditions (proper temperature, pressure and pH) may lead to impurities.

→ e.g → Improper heating (failing to achieve bright red temperatures) in process of manufacture of zinc oxide can lead to an oxidised metallic Zn as an impurity.

⑦ During Purification and Processing :-

• Often if not properly controlled, impurities also get added during the purification processes mainly through the purifying reagents, solvents or vessels used.

⑨ Reagents used to remove other impurities :-

→ Sometimes some chemicals are added to remove or to precipitate another substance. This may be also give rise to source of impurity.

→ For e.g $BaCl_2$ is added to remove excess of sulphate in $AlCl_3$, hence $AlCl_3$ is likely to contain Ba as an impurity.

(b) Solvents used in Purification :-

→ Often the solvents used for purification can be sources of impurities. These solvents range from organic solvents to acids (organic as well as mineral) and of course water.

(c) Contamination due to vessels and equipment used for purification :-

→ During the purification processes, if the vessels are defective or not perfectly cleaned and dried they may add impurity like metallic ions, rust, glass particles, moisture etc.

→ The other equipment mainly the filters, centrifuges, dryers etc, also need to be clean and dry.

(3) During storage and Packaging :-

(a) Errors in the Packaging :-

→ Similar looking products, such as tablets of the same size, shape and colour, packed in similar containers can result in mislabeling of either or both of the products.

(b) Microbial contamination :-

→ Microbial contamination, mainly in the form of fungal and bacterial, growth may be due to the result of improper storage conditions as well as faulty packaging.

→ The products for parenteral administration and opthalmic preparations have to undergo sterility tests.

Limit Test for Chlorides

Apparatus Required

Nessler cylinders
Glass rod
Stand

Chemicals Required

Dilute nitric acid (10%)
Silver nitrate (5%)
Sodium chloride

- **Reaction:-**

$$Cl^- + AgNO_3 \xrightarrow{dil\ HNO_3} AgCl\downarrow + NO_3^-$$

- **Principle:-**

It is based upon the chemical reaction between silver nitrate and soluble chloride in the presence of dilute nitric acid to give opalescence of silver chloride. The opalescence produced is compared with the standard solution.

→ If the opalescence in the sample is less than the standard, it passes the test.

→ If it is more the standard, it fails the test.

- **Procedure:-**

Take two same Nessler cylinders. Label one as "Test" and the other as "standard".

Standard	Test
(1) Place 1 ml of 0.0584% w/v solution of NaCl in a Nessler cylinder.	Dissolve the specified quantity of the substance in distilled water and transfer to Nessler cylinder.
(2) Add 10 ml of dil. HNO_3.	Add 10 ml of dil. HNO_3
(3) Dilute to 50ml with water and add 1 ml of silver nitrate soln.	Dilute to 50 ml with water and add 1 ml of silver nitrate soln.
(4) stir immediately with a glass rod and allow to stand for 5 minutes.	stir immediately with a glass rod and allow to stand for 5 minutes.
(5) observe the opalescence developed and compare with that of the sample.	observe the opalescence developed and compare with that of the sample.

Limit Test for Sulphatis

- **Apparatus Required :-**

Nissler cylinders, Glass rod, stand.

- **Chemicals Required :-**

(1) Dilute hydrochloric acid.

(2) 0.5 M Barium chloride - 122.1 g of $BaCl_2 . 2H_2O$ dissolved in distilled water.

(3) Barium sulphate reagent containing 0.5 M barium chloride in 1000 ml of water.

- **Reaction :-**

$$SO_4^{2-} + BaCl_2 \xrightarrow{dil\ H_2SO_4} BaSO_4 \downarrow + 2Cl^-$$

- **Principle :-**

It is based upon the chemical reaction between barium chloride and soluble sulphate in the presence of dilute hydrochloric acid. The turbidity produced is compared with the standard solution. Barium chloride reagent contains barium chloride, sulphate-free alcohol and small quantity of potassium sulphate. The inclusion of the small quantity of potassium sulphate in the reagent increases the sensitivity of the test.

Alcohol prevents super saturation and a more uniform turbidity develops.

→ If the turbidity produced in the test is more intense than the standard turbidity, then the drug fails the test. otherwise, it passes the test.

• Procedure :-

Take two 50ml Nessler cylinders. Label one as "Test" and the other as "standard".

Standard	Test
(1) Place 1 ml of 0.1089 %. w/v solⁿ of K_2SO_4 in a Nessler cylinder.	→ Dissolve the specified quantity of the substance in distilled water and transfer to Nessler cylinder.
(2) Add 2 ml of dil. HCl.	→ Add 2 ml of dil. HCl.
(3) Dilute to 45 ml with water and add 5ml of barium sulphate reagent.	→ Dilute to 45 ml with water and add 5ml of barium sulphate reagent.
(4) Stir immediately with a glass rod and allow to stand for 5 minutes.	→ Stir immediately with a glass rod and allow to stand for 5 minutes.
(5) Observe the turbidity developed and compare with that of the sample.	→ Observe the turbidity developed and compare with that of the standard.

Limit Test for Iron

- **Apparatus Required:-**

Nessler's cylinder, glass rod, stand.

- **Chemicals Required:-**

90% iron free citric acid solution, standard iron solution, thioglycolic acid.

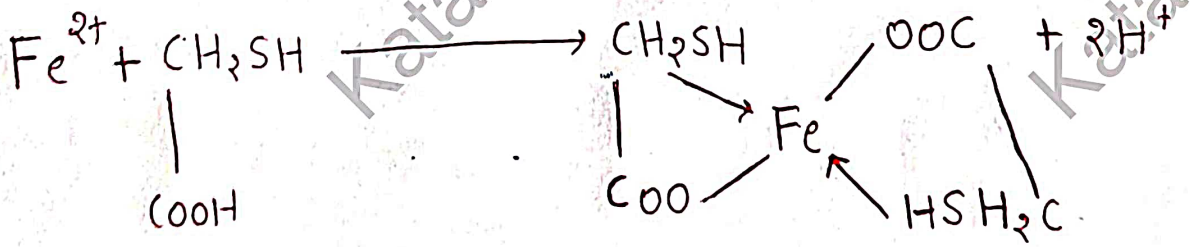
- **Principle:-**

The test depends upon the reaction between ferrous iron and thioglycollic acid in the presence of ammonia.
A pale pink to dup ruddish purple colour is produced.
Ferric iron is produced to ferrous ion by the thio-glycollic acid and the compound produced is ferrous thioglycollate.

→ Ferrous thioglycollate complex is colorless in acidic or neutral solutions.

→ only in the presence of alkali pale pink color will be produced.

- **Reactions :-**



Thioglycolic acid

ferrous thioglycolate.

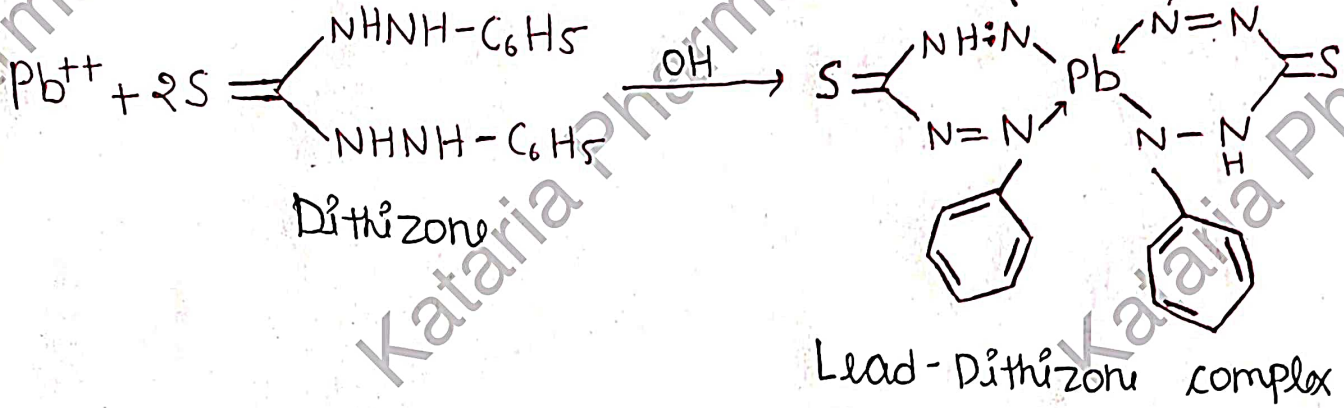
• Procedure :-

Standard	Test
(1) 2 ml of standard solution of iron diluted with water upto 40 ml.	Sample is dissolved in specific amount of water and then volume is made upto 40 ml.
(2) Add 2 ml of 20% w/v of citric acid (iron free).	Add 2 ml of 20% w/v of citric acid (iron free).
(3) Add 2 drops of thioglycollic acid.	Add 2 drops of thioglycollic acid.
(4) Add ammonia to make the solution alkaline and adjust the volume to 50 ml.	Add ammonia to make the solution alkaline and adjust the volume to 50 ml.
(5) Keep aside for 5 minute.	Keep aside for 5 minute.
(6) Color developed is viewed vertically and compared with standard solution.	Colour developed is viewed vertically and compared with standard solution.

Limit Test For Lead

Lead is a most undesirable impurity in medical compounds and comes through use of sulphuric acid, lead lined apparatus and glass bottles use for storage of chemicals.

• Reaction :-



Dithizone

Lead - Dithizone complex

• Principle :-

Limit test of lead is based on the reaction of lead and diphenyl thiocabazone (dithizone) in alkalin solution to form lead dithizone complex which is red in color.

→ Dithizone is green in color in chloroform and lead-dithizone complex is violet in color, so the resulting color at the end of process is red.

• Procedure :-

Standard	Test
(1) A standard lead soln is prepared equivalent to the amount of lead permitted in the sample under examination.	A known quantity of the sample solution is transferred in a separating funnel.

(2) Add 6 ml of ammonium citrate	→ Add 6 ml of ammonium citrate.
(3) Add 2 ml of Potassium cyanide and 2 ml of hydroxylamine hydrochloride.	→ Add 2 ml of Potassium cyanide and 2 ml of hydroxylamine hydrochloride.
(4) Make solution alkaline by adding ammonia solution.	→ Make solution alkaline by adding ammonia solution.
(5) Extract with 5 ml of dithizone in chloroform solution until it becomes green.	→ Extract with 5 ml of dithizone in chloroform solution until it becomes green.
(6) Dithizone extracts are shaken for 30 minutes with 30 ml of nitric acid and the chloroform layer is discarded.	→ Dithizone extracts are shaken for 30 minutes with 30 ml of nitric acid and the chloroform layer is discarded.
(7) To the acid solution add 5 ml of standard dithizone solution.	→ To the acid solution add 5 ml of standard dithizone solution.
(8) Add 4 ml of ammonium cyanide.	→ Add 4 ml of ammonium cyanide.
(9) Shake for 30 minutes.	→ Shake for 30 minutes.
(10) observe the colour.	→ observe the colour.

Limit Test for Heavy Metals

It is a limit test of the quantity of heavy metals contained as impurities in drugs. The heavy metals are the metallic inclusions that are darkened with sodium sulphide in acidic solution or hydrogen sulphide saturated solution as their quantity is expressed in terms of the quantity of lead (Pb).

- **Reaction:-**

$Pb(NO_3)_2$ (Pb^{+}) + H_2S ⟶ PbS↓ + 2HNO_3

$PbCl_2$ + Na_2S ⟶ PbS↓ + 2NaCl

- **Principle:-**

It is based on the reaction between the solution of heavy metals and a saturated solution of hydrogen sulphide. In acidic media, it produces reddish/black colour with hydrogen sulphide which is compared with the standard lead nitrate solution.

- **Procedure:-**

Standard	Test
(1) 2 ml of standard lead solution is taken in a Nessler cylinder and diluted to 25 ml with water.	Dissolve the specified quantity of the substance in distilled water diluted to 25 ml with water and transfer to Nessler cylinder.

(2) Adjust the pH to 3-4 by dilute acetic acid or dilute ammonia solution.	Adjust the pH to 3-4 by dilute acetic acid or dilute ammonia solution.
(3) Dilute further to 35 ml with water.	Dilute further to 35 ml with water.
(4) Add 10 ml of freshly Prepared H_2S solution.	Add 10 ml of freshly Prepared H_2S solution.
(5) Dilute to 50 ml with water.	Dilute to 50 ml with water.
(6) Mix and allow to stand for five minutes.	Mix and allow to stand for five minutes.
(7) observe the quantity of the black ppt of lead sulphide formed and compare with that of the standard.	observe the quantity of the black ppt of lead sulphide formed and compare with that of the standard.

Limit Test of Arsenic

• Principle :-

Limit test of Arsenic is basid on the reaction of arsenic gas with hydrogen ion to form yellow stain on mercuric chloride paper in presence of reducing agents like potassium iodide. It is also called as Gutzeit test and requires special apparatus.

Arsenic present as arsenic acid in the sample is reduced to arsenious acid by reducing agents like potassium iodide, stannous acid, zinc, hydrochloric acid. etc. Arsenious acid is further reduced to arsin (gas) by hydrogen and reacts with mercuric chloride paper to give a yellow stain.

• Reaction :-

$$H_3ASO_4 + H_2SnO_2 \longrightarrow H_3AsO_3 + H_2SnO_3$$

Arsenic acid — Arsenious acid.

$$H_3AsO_3 + 3H_2 \longrightarrow AsH_3 + 3H_2O$$

Arsin gas

$$2AsH_2 + HgCl_2 \longrightarrow Hg\begin{matrix}AsH_2\\AsH_2\end{matrix} + 2HCl$$

Mercuric chloride — Yellow colour

→ The dipth of yellow stain on mercuric chloride paper will depend upon the quality of arsenic present in the sample.

• Procedure :-

Standard	Test
(1) A known amount of dilute arsenic solution is kept in the wide mouthed bottle of the apparatus.	Dissolving specific amount of sample in water and stannous HCl (as free) and kept in the wide mouthed bottle of the apparatus.
(2) To this solution, 1 gm of KI, 5 ml of stannous chloride and 10 gm of zinc is added (all these reagents should be arsenic free).	To this solution, 1 gm of KI, 5 ml of stannous chloride and 10 gm of zinc is added (all these reagents should be arsenic free).
(3) Kep the solution aside for 40 minutes.	Kep the solution aside for 40 minutes.
(4) Compare the stain obtained on the mercuric chloride paper with that in the apparatus containing test solution.	Compare the stain obtained on the mercuric chloride paper with that in the apparatus containing test solution.

Modified Limit test for chlorides

• Apparatus required :-

Nessler's cylinder, measuring cylinder, pipette, spatula.

• Chemicals required :-

Distilled water, dilute nitric acid, 0.1 M silver nitrate solution, Potassium permanganate sample.

• Principle :-

→ The limit test for chloride based on the reaction between soluble chloride and silver nitrate to give white precipitate of silver chloride. This white precipitate are insoluble in dilute nitric acid and give turbidity or opalescence to the test solution. If the turbidity developed in the sample is less than the standard turbidity, the sample passes the limit test for chloride and vice-versa.

→ As potassium permanganate gives purple color aqueous solution that interfere in the comparison of opalescence or turbidity, therefore the aqueous solution must first be decolorized. Potassium permanganate is oxidizing agent while ethanol is reducing agent.

→ when potassium permanganate solution is treated with ethanol in presence of heat the redox reaction will take place, i.e. potassium permanganate gets reduced to manganese di-oxide. The filtrate of the reaction is colorless that is subjected to proceed for limit test for chloride.

• Chemical reaction :-

$$2KMnO_4 + 3C_2H_5OH \longrightarrow 2MnO_2 + 2KOH + 2CH_3CHO + 2H_2O$$

$$Cl + AgNO_3 \xrightarrow{HNO_3} AgCl + NO_3.$$

• Procedure :-

Standard	Test
(1) Take 10ml chloride standard solution (25 PPM chloride) and add 5 ml water in a Nessler's cylinder.	Transfer the prepared test solution in Nessler's cylinder.
(2) Add 10 ml of dilute nitric acid and dilute to 50 ml with distilled water.	Add 10 ml of dilute nitric acid and dilute to 50 ml with distilled water.
(3) Add 1 ml of 0.1 M silver nitrate solution and stir immediately with glass rod and allow standing for 5 minutes protected from light.	Add 1 ml of 0.1M silver nitrate solution and stir immediately with glass rod and allow standing for 5 minutes protected from light.

→ Compare the turbidity or opalescence produced in test solution with standard solution and report the result.

Modified Limit test for Sulphate

• Principle :-

→ It is a comparison method. It involves the comparison of opalescence or turbidity of test sample versus standard sample which contain sulphate impurities.

→ The limit test of sulphate is performed on the basis of reaction between the barium chloride reagent and soluble sulphate in the sample with formation of barium sulphate ($BaSO_4$) white precipitates.

→ Sulphate free alcoholic potassium sulphate is added to increase the sensitivity of the test. very small amount of barium sulphate present in the reagents acts as a seeding agents for precipitation of barium sulphate. If sulphate is present in the sample under the test.

• Ethanol is added to prevent the super saturation i.e the crystallization of sulphate with any other ion.

• Reactions :-

$$K_2SO_4 + BaCl_2 \xrightarrow{\text{Acetic acid}} BaSO_4 + -2KCl$$

$$SO_4^{2} + BaCl_2 \xrightarrow{\text{Acetic acid}} BaSO_4 + 2Cl^-$$

$$2KMnO_4 + 3C_2H_5OH \longrightarrow 3CH_3CHO + 2KOH + 2MnO_2 + 2H_2O$$

• Procedure :-

• Preparation of Test solution ($KMnO_4$) :-

Dissolve 1.5g in some of distilled water, heat on a water-bath and add gradually 6ml of ethanol (95%). Cool, dilute to 60ml with distilled water and filter.

Take two 50ml Nessler cylinders. Label one as "Test" and the other as "standard".

Standard	Test
(1) Mix 15 ml of sulphate standard solution and 15 ml of distilled water in a Nessler cylinder.	Take 10ml of the above test solution in Nessler cylinder.
(2) Add 0.15 ml of 5N acetic acid.	Add 15ml of 5M acetic acid.
(3) Add 2.5 ml barium sulphate reagent.	Add 2.5 ml barium sulphate reagent.
(4) Add sufficient distilled water to produce 50 ml.	Add sufficient distilled water to produce 50 ml.
(5) Stir immediately with glass rod and allow to stand for 5 minutes protected from light	Stir immediately with glass rod and allow to stand for 5 minutes protected from light.

Compare the turbidity or opalescence in the test solution by viewing transversely both solutions against black background.