

# DRUG EVALUATION

# DRUG EVALUATION

Drug evaluation involves the determination of identity, purity and quality of a drug.

**Identity** – identification of exact authentic biological source of the drug

**Purity** – absence of foreign material either organic or inorganic

**Quality** – concentration of therapeutically active constituents

**ADULTERATION** – incorporation of impurities, removal of all or part of the active constituents of a drug.

Addition of low grade and spoiled drugs or entirely different material similar to that of genuine drug substituted with an intention to cheat.

Adulteration may be defined as a admixture or substitution of genuine drugs with spurious , inferior, defective or otherwise useless or harmful substances

Adulteration involves

**Deterioration** : impairment in drug quality

**Admixture** : addition of objects due to carelessness or ignorance

**Sophistication** : intentional type of adulteration

**Substitution** : addition of an entirely different substance instead of the original drug

**Inferiority** : Any sub standard drug

**Spoilage** : occurs as a result of microbial attack

# TYPES OF ADULTERANTS

## Adulteration with sub standard commercial varieties

Which resembles the original crude drug but they are sub standard in nature and of low cost – (for eg) *strychnous nuxvomica* is adulterated with *strychnous Nux blanda* or *strychnous potatorum*.

- *Capsicum minimum* is adulterated with *capsicum annum*
- Indian senna is adulterated with Arabian senna and Dog senna
- Gentian is adulterated with kutki etc

## Adulteration with superficially similar inferior drugs

These inferior drugs either have or do not have the chemical or therapeutic characters of the original crude drug, morphologically they resemble the original drug (for eg) belladonna leaves are adulterated with ailanthus leaves

- Saffron is adulterated with *Carthamus tinctorius*
- Myrrh is adulterated with scented *bdellium* etc

## **Adulteration with Artificially manufactured substances**

Some substances are artificially prepared to make them look similar to the original drug . ( For eg ) coffee is adulterated with compressed chicory

- Bees wax is adulterated with yellow coloured paraffin wax
- Nutmeg is adulterated with properly cut and shaped basswood etc

## **Adulteration with exhausted drugs**

These drugs are similar to the original drug but active constituents are taken out or extracted out

- ( For eg ) Fennel, clove , coriander , caraway etc
- Some times additives are added to the exhausted drugs to modify their taste and colour ( for eg ) aloes are added to make the exhausted gentian bitter
- Exhausted saffron is coloured by adding artificial colours

## **Presence of vegetative matter from same plant**

Some small plants similar to the medicinal plants in their colour , odour and constituents are mixed with the original drug.

( for eg ) lower plants like moss, liver worts are used for adulterating cascara or cinchona those growing on stem are mixed with stramonium , lobelia and senna( leaf drugs )

## **Harmful Adulterants**

Mixing of waste materials with the original drugs especially in case of liquids or unorganized drugs ( for eg ) amber coloured glass pieces for adulterating colophony , limestones for asafoetida, lead shot for opium , white oil for coconut oil , cocoa butter for stearin or paraffin etc

## **Adulteration with non plant materials**

Clove with clay , benzyl benzoate with balsam of peru, citral to citrus oils like oil of lemon and orange etc

## **Adulteration of powders**

The chances of adulteration in powdered drugs are more in comparison to the entire drug

( for eg ) Dextrin for adulterating ipecacuanha,

- Powdered olive stones for powdered liquorice or gentian
- Exhausted ginger powder for powdered colocynth or ginger
- Red sanders wood for capsicum
- Brick powder is often used for bark drugs

# EVALUATION

- ORGANOLEPTIC EVALUATION
- MORPHOLOGICAL EVALUATION
- MICROSCOPIC EVALUATION
- PHYSICAL EVALUATION
- CHEMICAL EVALUATION
- BIOLOGICAL EVALUATION



## **ORGANOLEPTIC EVALUATION**

Drug evaluation by means of our organs of sense and it includes gross morphology and other sensory characters like odour, colour, taste and texture

## **MORPHOLOGICAL EVALUATION**

The macroscopy of a drug means its external appearance like parts of plants

**BARKS** : All the tissues as a woody stem. Barks are collected by stripping from the trunk or branches of appropriate tree.

( for eg ) cinnamon, cinchona, quillia, asoka and kurchi bark etc

During drying process un equal contractions cause the dried bark in to different shapes like flat , curved , re curved, channeled , quill, double quill and compound quill

**UNDER GROUND STRUCTURES** : Roots, Rhizomes and stolons

**Roots** have no buds , scale leaves or leaf scars and have a central core of woody xylem tissue . ( for eg ) podophyllum , liquorice , jatamansi , Rauwolfia

**Rhizomes** are underground stems which have buds and scale leaves and scars.

They have a central pith and the drugs include ginger, turmeric, dioscorea

Different shapes are like cylindrical , conical , fusi form, straight, tortous

**Leaves** : there are many different shapes and sizes of leaves. One leaf is comprised of all the photo synthetic organs arising from a node on a stem.

This is called as a simple leaf, some times one leaf may consist of many leaf lets it is termed as a compound leaf

Different shapes of leaves , their margin , base, apex and venation are helps for the identification of drugs.

**Shapes** – Acicular, elliptical , oval , oblong, round , linear, lanceolate, ovate, obovate, cordate ,reniform.

Margins – entire, serrate, dentate, crenate, sinuate

Apex – emarginate, re curved , obtuse, acute , acuminate, mucronate

Bases – assymmetric , cordate , segitate , decurrent

Venation – parallel , reticulate palmate, reticulate pinnate

**FLOWERS** : These are the re productive organs of a plant and consists of four basic parts calyx, corolla, androecium , and gynoecium. Many plants have inflorescence. Group of flowers from single stalk.

( for eg ) solitary,cymose,raceme,spike,corymb, simple umbel, compound umbel

**FRUITS** : Fruits are the organs of the plant containing seeds,they arise from the ovary and sometimes from other parts of the flower.

( for eg ) cardamom,colocynth and bael.

The shapes are globular, oblong, or ellipsoidal

**SEEDS** : Seeds arise from the ovules in the carpels of the flowers and are characterized by the hilum , micropyle , raphe .

( for eg ) isphaghul, linseed, nux vomica, psoralea

The shapes of seed may be of different types like globular , oval , reni form , plano convex and spherical

**HERBS** : A full description of a herb includes stem, leaves, flowers, and fruits

**STUDY OF SENSORY CHARACTERS** : observation of **colour, taste and smell.**

The drugs if they are dried in sun loses its colour, and smell . Such drugs should be shade dried . Capsicum, ginger has pungent taste. Gentian and chirata have bitter taste. Glycyrrhiza ( liquorice) has sweet taste.

Deterioration of drugs like ergot or cantharides due to improper storage emit ammonical odour.

# MICROSCOPIC EVALUATION

- Sections of crude drugs are taken and after mounting and staining it can be examined under microscope.
- Transverse or longitudinal sections were taken and the arrangement of tissues are observed under microscope.
- The basic layout of tissues in stems, roots, leaves etc .
- The cells are identified like fibres, sclereids, tracheids, vessels and cork .
- The cell contents include starch, crystals of calcium oxalate and fixed oil can be observed.
- In the powdered drugs thick walled cells, lignified cells with lignified walls can be observed.
- Important features of the cells can be drawn with the help of the camera lucida

## PHYSICAL EVALUATION

- Physical constants such as viscosity of drugs containing gums
- Swelling factor of the mucilage , Foam content of the saponin drugs,
- Determination of the extractive values and ash values and volatile oil content can be determined .
- Physical constants like boiling point, freezing point, absorption coefficient ( ability of the substance to absorb some specific compounds )
- Refractive index, optical rotation, spectroscopic analysis, fluorescence analysis and radio immune assays of drugs such as alkaloids, volatile oils , fixed oils can be determined.

## FOREIGN MATTER

- Crude drugs has pharmacopoeial limit for the impurities such as animal excreta , insects or moulds.
- 25-100gm of powdered drug is taken and spread in thin layer on a paper.
- The foreign matter is picked up and weighed. If the percentage exceeds pharmacopoeial limits the sample is declared sub standard.
- Some times the crude drugs get contaminated with certain microbes like bacteria , fungi and moulds. Such contaminations can lead to certain toxicities.
- Under such circumstances the drugs may have to be subjected to sterilization in special equipment by treatment with ethylene oxide.

# MOISTURE CONTENT

- Presence of moisture in a crude drug can lead to its deterioration due to either activation of certain enzymes or growth of microbes
- Moisture content can be determined by heating the drug at 105 C to constant weight and calculating the loss of weight.
- Moisture in the crude drug can also be determined by distilling the drug in presence of water immiscible solvent like toluene or xylene and from the distillate thus collected level of water can be measured.
- Karl fisher method is a standard procedure for determining moisture content.



# EXTRACTIVE VALUES

For determining the active chemical constituents water, alcohol or ether soluble extractive values are determined.

- Water soluble extractive values in case of drugs like aloe, gentian and liquorice
- Alcohol soluble extractive values in case of drugs like ginger, valerian and quillia.
- All such extracts are prepared in an appropriate solvent either by maceration or by continuous extraction process.

**VOLATILE CONTENT** : Volatile oil containing drugs like cardamom, cinnamon, clove, fennel, dill etc depends upon the amount of the volatile content present in the drug. Weighed quantity of the drug is boiled with water

- In a round bottom flask fitted with clevengers apparatus, the distillate is collected in graduated tube volatile oil is lighter than water remains on the top and the amount thus collected is read from the tube.

**ASH VALUES** : The total ash content is determined by incinerating a known quantity of the air dried crude drug in a silica crucible and the residual ash is weighed to calculate the percentage of ash on the basis of initial dry weight of the drug sample. Very high ash values indicate the contamination.

**REFRACTIVE INDEX** : For the evaluation of volatile and fixed oils measurements of refractive index is of significance.

- The refractive index changes if a particular oil is mixed with another oil.
- The refractive index is defined as the ratio of the velocity of light in vacuum to velocity in the substance and is ratio of the angle of incidence to the angle of refraction.
- It is measured by refractometer.

# OPTICAL ROTATION

- Many substances of biological origin can rotate the plane of polarised light either to right or to the left side.
- Such compounds are said to be optically active and are designated as dextro rotatory or levo rotatory as the beam is deflected to the right or the left respectively.
- The rotation is expressed in degrees plus ( +) indicating rotation to the right, minus (-) indicating deflection to the left.
- A Polarimeter is used to measure optical rotation .
- Most volatile oils contain optically active components and the direction of the rotation as well as its magnitude is a useful criteria of purity.

# RF VALUES

- Thin layer chromatography ( TLC) has become increasingly popular for both qualitative and quantitative evaluation of drugs.
- Rf value refers to the ratio of distance moved by the solute to the distance moved by the solvent on a thin layer of an adsorbent.
- Rf value of the component is characteristic and can be used to identify the component by comparison with the reference standard.
- The intensity of the colour of the spot of the compound under test can be utilized for quantitative estimation of the principle in the drug.

$$R_f = \frac{\text{Distance travelled by the solute( component)}}{\text{Distance travelled by the solvent front}}$$

# SPECTROSCOPIC ANALYSIS

- Specific groups of phyto constituents are reported to be present in crude drugs and thus assay of a particular group of phyto constituents

( for example) glycosides of a cardio tonic drug or alkaloids of a solanaceous drug helps in the evaluation of drugs.

- In spectroscopic analysis the capacity of certain molecules of a drug to absorb vibration at specific wave length is the basis for the drug evaluation
- Most groups of active phytoconstituents of drugs are determined by spectrometry including colorimetry and fluorescence analysis.
- Quinine sulphate in aqueous solution can be determined by a spectro fluorometer.

# CHEMICAL EVALUATION

- Determination of active constituent in a drug by chemical process is referred to as chemical evaluation.
- **Vitali morin reaction** is done for determining tropane alkaloids in datura, Belladonna and stramonium.
- **Borntragers test** is used for the detection of anthraquinone glycosides in purgative drugs like senna, rhubarb, cascara and aloe .
- Acid value, iodine value, saponification value , ester value etc are employed for fixed oils and fats.
- Evaluation of alkaloidal containing drugs can be done by determining total alkaloidal content through acid base titration or by non aqueous titrations of alkaloidal salts.
- Chemical evaluation of crude drugs used for the identification as well as estimation of the phyto constituents present in a drug.

# BIOLOGICAL EVALUATION

- The drug is administered to animals and the response on a particular organ or tissue or the whole animal is measured and compared with the standard drug under the same set of conditions.
- Drug containing cardiac glycosides can be biologically evaluated on cats , frogs or pigeons .
- The bitter drugs like quassia, gentian and chirata can be evaluated by the bitter value.
- Ginger and capsicum can be evaluated for their pungency.
- Anthelmintic drugs like malefern can be evaluated by their action on earth worms.
- The biological evaluation can be done either on an isolated tissue or on an intact animal.

- Biological evaluation is performed using whole animals, animal preparations, isolated living tissues or micro organisms.
- This assay method is termed biological assay or bio assay as living organisms used.
- This method is less precise, more time taking and expensive.
- Herbal drugs are standardized by the methods evaluating their biological efficacy
- Hepato protective activity, hypo glycemc activity, Anti fertility activity, Anti inflammatory activity, Neuro pharmacological activity.

### **MICROBIOLOGICAL ASSAY**

1. Cylinder plate method
2. Turbidimetric method



## **CYLINDER PLATE METHOD**

- A solid nutrient medium is poured in petri plates
- And inoculated with a microbial culture. Cylindrical holes are made of suitable sizes in the medium surface and test compound of various dilutions are poured in them.
- After incubation the diameter of microbial growth inhibition surrounding the cylinders is measured, inhibition produced by the test compound is compared with the standard.

## **TURBIDIMETRIC METHOD**

- In this method microbial growth inhibition indicated by turbidity of microbial suspensions in a fluid medium containing the test compound is measured .
- Transmittance changes produced by the test compound and those produced by the standard of known concentrations are compared.