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DRUG ADULTERATION

An adulterated drug means one which does not conform to the official requirements. Adulteration involves incorporation of impurities. spoilage, deterioration, admixture, sophistication and substitution. The genuine drugs are substituted with spurious, inferior, defective or harmful substances. The spoiled or deteriorated drugs represent the greatest percentage of drug adulteration. In some cases the dealers substitute the drugs with cheap materials in case of scarcity or when the price of a drug is high. The adulteration may be due to faulty collection, imperfect perparation and incorrect storage as described hereunder :

FAULTY COLLECTION : In some cases the proportion of medicinally-active constituent reaches a maximum at a particular season, stage of development, or age. But collection of correct part of genuine plant without regard to time factors causes adulteration. The following are some examples:

(i) Season

Drug	**Season of Maximum Activity**
Solanaceous leaves	Flowering stage of the drug (Summer)
Wild Cherry bark	Autumn
Colchicum corm	Early summer
Male fern	Late autumn.

INCORRECT STORAGE : Incorrect storage spoils many drugs. The quality, value or usefulness of the drug has been impaired or destroyed by the action of moisture, light, temperature and microorganisms (fungi and bacteria) and the article becomes unfit for human consumption. Many examples of spoilage are found in food industry. All drugs which are unfit for human or animal consumption are legally considered as adulterated. The impairment of the quality or value of an article by the abstraction or destruction of valuable constituents by distillation, extraction, aging, moisture, heat, fungi, insects or other means deteriorate the drugs considerably. A few examples are :

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EVALUATION OF DRUGS

Evaluation of drugs deals with the correct identification of the plant and determination of quality and purity of the crude drugs. Actual collection of the drug is done from the identified plant or animal. For this purpose research gardens have been maintained. The characters of an unknown sample are compared with the authentic monographs written in the pharmacopoeia. The high quality of the drug is maintained by collection of the drug from the correct natural source at proper time; preparation of samples of the collected drugs by proper cleaning, drying and to free from dirt, and proper preservation of the cleaned, dried and pure drug.

The evaluation of a drug is done by studying its organoleptic, microscopic, biological, chemical, and physical properties.

ORGANOLEPTIC EVALUATION

Organoleptic evaluation means study of a drug with the help of organs of sense which includes its external morphology, colour, odour, taste, sound of its fracture, etc.

Morphological Characters : To study morphology of a drug, its shape and size, colour and external markings, fracture and internal colour, odour and taste are examined. The organized drugs are classified into :

1. **Barks :** Which are tissues in a woody stem outside the inner fascicular cambium, e.g., Cinnamon, Cinchona, Quillaia, Ashoka and Kurchi.

2. **Underground Structures** : Which may be rhizomes, roots, bulbs, corm, and tubers; they are often swollen due to storage of carbohydrates and other chemicals, e.g., roots (Podophyllum, Liquorice, Jatamansi, Rauwolfia), rhizomes and stolons which are underground stems and have buds, scale leaves and scars, (Ginger, Turmeric, Dioscorea).
3. **Leaves** : These are photosynthetic organs arising from a node on a stem. The shape, margin, base, apex and venation of leaves help in the identification of the drugs. Senna, Tulsi, Vasaka and Digitalis leaves can be easily identified.
4. **Flowers** : These are reproductive organs of a plant and possess different shapes, size and colour, e.g., Saffron, Banafsha, Pyrethrum.
5. **Fruits** : Fruits arise from the ovary and contain seeds, e.g. Cardamom, Colocynth, Almond, Vidang, Bahera, Amla and Bael.
6. **Seeds** : Seeds are developed from the ovules in carpels of the flowers and characterized by the hilum, micropyle and sometimes raphe. The seed drugs are Ispaghula, Linseed, Nux-vomica, Psoralia.
7. **Herbs** : The whole aerial part is sometimes used as a drug, e.g. Brahmi, Chirata, Kalmegh, Pudina, Shankhpushpi, etc.

The shape of a drug may be cylindrical (Sarsaparilla), sub-cylindrical (Podophyllum), conical (Aconite); fusiform, ovoid or pyriform (Jalap), and terete or disk-shaped (Nux-vomica). The drug may be simple, branched, curved or twisted. The length, breadth and diameter are measured in millimeters or centimeters. In case of conical drugs the size of both parts is mentioned.

External markings are mentioned as :

1. furrows, ridges, etc.,
2. wrinkles,
3. annulations,
4. fissures,
5. nodules,
6. projections,
7. scars of leaf, stem-base, root, bud, bud-scale, etc.

The fractures may be complete, incomplete, short, fibrous, splintery (breaking irregularly), brittle (easily broken), tough and weak.

Sensory Characters : Colour, texture, odour and taste are useful in the evaluation of drugs. This method is especially applicable to drugs containing volatile oils or pungent principles (e.g. Capsicum), and to the detection of the effects of inadequate drying or damp storage. The external colour varies from white to yellowish grey, brown, orange or brownish black. The colour of some drugs changes if they are dried in sunlight in place of shade.

The odour of a drug may be either distinct (characterisic) or indistinct. The terms used to define odour are aromatic, balsamic, spicy, alliaceous (garlic-like), camphoraceous (camphor-like), terebinthinate (turpentine-like) and others. Leaves of different species of *Mentha* can be distinguished by smell. Clove and exhausted clove are differentiated by odour. Deteriorated Cantharides have ammonical smell while spoiled Ergot has rancid and ammonical smell.

Taste is a particular sensation production by certain substances when these come into contact with taste buds present in epithelial layer of the mouth. The taste may be sour (acidic), salty (saline), sweet (saccharine), bitter, alkaline and metallic. Substances possessing no taste are mentioned as tasteless. The tastes due to a characteristic odour are grouped as aromatic, balsamic, spicy, alliaceous, camphoraceous and terebinthinate. The taste produced by distinctive sensations to the tongue are classified as mucilaginous, oily, astringent (producing a contraction of the tissues of the mouth), pungent (warm biting sensation), acrid (unpleasant, irritating sensation) and nauseous (causing vomiting).

The drugs like Ginger and Capsicum have pungent taste; Gentian, Chirata and Kalmegh have bitter taste; Glycyrrhiza and Honey are sweet in taste. Linseed and Isphagula are mucilaginous; fixed oils have bland taste; calcium oxide is astringent; Podophyllum, Kaladana, Jalap and Ipomoea are acrid; while Ipecac, Acorus, and *Tylophora indica* contain nauseous taste.

Glycyrrhiza has hard and fibrous fracture due to the presence of fibrous and woody tissues. Aconite has a horny fracture due to gelatinization of starch.

Colour of drugs are standardized and determined by the Inter-Society Colour Council-National Bureau of Standard method. For example, reserpine is described as a "white or pale buff to slightly yellowish, odourless crystalline powder".

MICROSCOPIC OR ANATOMICAL EVALUATION

Schleiden (1847) used microscope for the examination of drugs. Microscopic examination of section and powder drugs, aided by stains, helps in distinction of anatomy in adulterants. Further, microscopical examination of epidermal trichomes and calcium oxalate crystals is extremely valuable, especially in powdered drugs. In the powdered drugs the cells are mostly broken, except lignified cells. The cell contents such as starch, calcium oxalate crystals, aleurone, etc. are scattered in the powder. Some fragments are specific for each powder which may consist of parts of cells or groups of cells.

Plant parts are made up of specific arranged tissues, spores (Lycopodium) or hairs (Lupulin). Histological characters are studied from very thin transverse, or longitudinal sections properly mounted in suitable stains, reagents or mounting media.

The size, shape and relative positions of the different cells and tissues, chemical nature of the cell walls and of the cell contents are determined. The basic arrangement of tissues in each drug is fairly constant. Fibres, sclereids, tracheids, vessels and cork are least affected by drying. Starch, calcium oxalate, epidermal trichomes and lignin are examined carefully.

Microscope is also used for a quantitative evaluation of drugs and adulterated powders. This is done by counting a specific histological feature such as stomatal index, vein-islets and vein termination numbers, palisade ratio, etc. These features are compared with the standard samples.

Palisade Ratio : The average number of palisade cells beneath each epidermal cell is called as palisade ratio. It is determined from powdered drugs with the help of camera lucida.

Stomatal Number : The average number of stomata per square millimeter of the epidermis is known as stomatal number. The range and average value for each surface are recorded.

Stomatal Index : The percentage proportion of the number of stomata form to the total number of epidermal cells of a leaf is termed the stomatal index :

S.I. = S/E+S x 100; where S = number of stomata per unit area, E = number of ordinary epidermal cells in the same unit area.

Stomatal number varies considerably with the age of the leaf but the stomatal index is highly constant for a given species.

Vein-Islet Number : The word 'Vein-islet' is used for the minute area of photosynthetic tissue encircled by the ultimate divisions of the conducting strands. *Vein-islet number* is defined as the number of vein-islets per square mm calculated from four contiguous square mm in the central part of the lamina, midway between the midrib and the margin. The average range of vein-islet numbers for Senna are : *Cassia senna* (26), *C. angustifolia* (21); for Coca: *Erythroxylum coca* (11), *E. truxillense* (20); for Digitalis. *Digitalis purpurea* (3.5) *D. lanata* (2.7); *D. lutea* (4.4), *D. thapsi* (1.2).

Veinlet Termination Number : It is defined as the number of veinlet terminations per mm^2 of leaf surface. A vein termination is the ultimate free termination of a veinlet or branch of a veinlet. By this character different Coca leaves and Senna leaflets are differentiated.

CHEMICAL EVALUATION

Chemical evaluation involves the determination of active constituents by a chemical process. Chemical tests are used to identify certain crude drugs to determine purity. Chemical tests for alkaloids, carbohydrates, steroids, phenolic compounds, saponins, proteins, amino acids, fixed oils and volatile oils are performed. Titrimetric assay, iodine value, saponification value, acid value, acetyl value, ester value, peroxide value, hydroxyl value and ash value are determined. Tropane alkaloids in Datura, Belladonna and Stramonium are determined by Vitali-Morin reaction. Potassium chlorate and hydrochloric acid are used to estimate emetine in Ipecac. Strychnine in Nux-vomica is detected with ammonium vanadate and sulphuric acid. Bornträger's test is useful for detecting anthraquinone glycosides, present in Senna, Rhubarb, Cascara and Aloe. Alkaloid contents can be evaluated by determining total alkaloidal contents by acid-base titration.

Preparation of an extract by an appropriate solvent is sometimes applied to determine the quality of drugs. The solvent may extract a single constituent, e.g. fixed oil from crushed Linseed. Further examples of the use of extractive tests are in cases of Gentian, Colocynth seeds, Indian hemp, Ginger, Calumba, Rhubarb, Glycyrrhiza and Myrrh.

Drugs containing volatile oils are examined for authenticity and quality by determining the percentage of volatile oil yielded by steam distillation in a suitable apparatus. Standards for content of volatile oil in drugs usually allow a somewhat smaller percentage from powdered drugs as compared with the whole drug due to inevitable loss on grinding, volatilization and decomposition.

On ignition of crude drugs a residue of mineral substances or ash remains, derived from the cell wall and cell contents. The ash value is useful in determining authenticity and purity of drugs. For a number of official drugs, a limit is placed on the yield of acid-insoluble ash, i.e. the ash remaining after extraction of the total ash with dilute acid. This residue consists chiefly of silica, partly derived from the constituents of the cells and their walls and partly from foreign mineral matters, mainly soil. Acid-insoluble ash limits are imposed especially in cases where foreign silica may be present or when the calcium oxalate

contents of the drug is high. Pharmacopoeial limits for acid insoluble ash vary from 0.5 (Agar) to 12 percent (Hyoscyamus). Glandular trichomes present in Hyoscyamus have a capacity of retaining clay and thus the acid insoluble ash value is higher in such cases. In case of Glycyrrhiza the total ash figure is of importance which indicates the care taken in the preparation of the drug. For the determination of total ash values the carbon must be removed below 450°C, since alkali chlorides would be lost due to volatile at high temperature. The total ash usually consists of carbonates, phosphates, silicates and silica. In case of Ginger a minimum percentage of water-soluble ash is determined to detect the presence of exhausted ginger.

PHYSICAL EVALUATION

Physical constants such as elasticity in fibres, viscosity of drugs containing gums, swelling factor of mucilage containing materials, froth number of saponin drugs, congealing point of volatile and fixed oils, melting and boiling points and water contents (loss on drying at 110°C) are some important parameters used in the evaluation of drugs. Ultraviolet light is also used for determining the fluorescence of extracts of some drugs (Gambir, Senna) and colours of alkaloids as : aconite (light blue), berberine (yellow), emetine (orange) and quinine (dense fluorescence in dilute sulphuric acid). The florescence of Belladonna leaf and root, Wild Cherry bark and Jalap is due to the presence of a coumarin, β-methyl asculetin. Pale Catechu shows fluorescence in alkaline solution due to gambir-fluorescin. Aloe exhibits a green fluorescence in a solution containing borax. Many other drugs show a marked intensity of colour or a characteristic colour under UV light. Rhubarb is differentiated from Rhapontic, Chinese or Indian Rhubarb by its marked fluorescence in UV light.

Physical constants are extensively applied to the active principles of drugs, such as alkaloids, volatile oils, fixed oils, etc. Solubility expresses number of ml of solvent require to dissolve one gram of the drug. For example, 1 g of codeine sulphate is soluble in 30 ml of water, and in 1300 ml of alcohol. Alkaloids and other nitrogenous compounds are soluble in dilute hydrochloric acid. Melting points are recorded for solid fixed oils (fats) and alkaloids.

Most of the monoterpenes have asymmetric carbon.

BIOLOGICAL EVALUATION

The drugs, which cannot be assayed satisfactorily by chemical or physical means, are evaluated by biological methods. Tests are carried out on intact animals, animal preparations, isolated living tissues or micro-organisms. Since living organisms are used, the assays are called 'biological assays'. Biological standardization procedures are generally less precise, more time consuming and more expensive to conduct than chemical assays. Therefore, they are generally used if the chemical identity of the active principle has not been fully elucidated; if, no adequate chemical assay has been derived for the active principle as in case of insulin; if the drug is composed of complex mixture and activity, e.g. Digitalis; if the purification of crude drug is not possible, e.g. separation of vitamin D from irridiated oils; and if the chemical assay is not a valid indication of biological activity.

A biological assay measures the actual biological activity of a given sample. In any one test the animals of only one strain are used. For some assays a specific sex must be used. The male rat has faster growth rate than the female. Therefore, use of both male and female in a growth test