IAMJ

Review Article International Ayurvedic Medical Journal ISSN:2320 5091

WHO (WORLD HEALTH ORGANIZATION) GUIDELINES FOR STANDARDIZATION OF HERBAL DRUGS

Nilakshi Pradhan[1], Jyoti Gavali[2], Nitin Waghmare[3]

[1]HOD, Shalakyatantra Department, [2]Associate Professor, Rasashastra Department,

[3]HOD, Kaumarbhritya, Associate Professor;

Shalakyatantra Department, Sumatibhai Shah Ayurved Mahavidyalaya, Pune, Maharshtra, India

ABSTRACT

Herbal drugs are being used as medicines from ancient period. The increased use of herbal drugs, and concerns over their safety and efficacy have certainly augmented the need of standardization of these herbal drugs. WHO has set up guidelines for standardization of these drugs, which are used as a standard by the majority of countries. The standardization includes the external (macroscopy/microscopy) as well as internal examination/ash values, extractive values and many other parameters to identify, authentify and study its chemical composition. Standardization of the medicinal plants will ensure indirectly that the plants are conserved for their medicinal and nutritive value. Standardization confirms the safety of the medicinal plant but efficacy has to be judged clinically or in the laboratory. There is a thin line between efficacy and the presence of chemical compounds in the drug. The major hurdle in standardization of the batch to batch variation in the plant compounds. Addition of finer analytical methods of the chemical compounds may help to minimize the variation and give a better resolution of the plant drug. Importance of toxicological examination has increased manifolds as contamination can occurs at various stages, from collection, storage, analysis or processing to extraction of active principles. These parameters should be recorded for years together; their database should be generated, recorded and analyzed statistically to see the difference in quality and quantity of the chemical compounds.

Keywords: Standardization, herbal drug, botanical, pharmacological, toxicological, parameters

INTRODUCTION

Medicinal plants are in use for the purpose of treatment of different ailments since centuries. There has been an immense increase in sales of herbal OTC (Over the counter drugs. This is growing to a billion dollar industry. The need for safety and efficacy has also escalated since the western interest has grown. Thus the need for standardization has come into view. The process of evaluation of the quality and purity of crude drugs by means of various parameters like morphological, microscopical, physical, chemical and biological observations is called standardization.[1]Standardisation of the herbal drug begins from the collection of the herbal drug to its packaging/use as medicine. The impediments in standardization of herbal drugs-

- Variability in the chemical composition of the soil and changes in the climate in-

fluence the range of phyto constituents present in the herbal drugs.[2]

- Growing deforestation is leading to increase in the number of endangered species of medicinal plants. This leads to addition of adulterants or substitutes to the herbal drug. Addition of adulterants and substitutes can change the safety and efficacy of the drug.

Objectives: The present article will review the WHO guidelines for standardization of Herbal Drugs as well as focus on the current and future trends of the methods used for analysis of the herbal drug.

Thus to overcome these obstacles there are many standards and parameters set by different pharmacopeias, but guidelines set by the WHO remain most significant.

Materials and Methods

BOTANICAL PARAMATERS[4]-

Sensory evaluation-Visual macroscopy, Colour, Odour, Taste, Fracture are the common tests conducted for identification of the crude drug.

Foreign matter-It has to be determined if the foreign matter is organic (Moulds, Insects, Animal excreta etc.) or Inorganic (Stone, soil etc).Foreign matter is considered as-

- material not collected from the original plant source (insects, moulds, or other animal contamination)
- Parts of the organ, or organs from which the drug is derived other than the parts named in the definition and description.

Methods to determine the foreign organic matter-

- Manual method
- Lycopodium spore method

Microscopy-Identification of histological characters(under low and high power).Study individual characteristics in all respects (Qualitative and Quantitative measurements).Observe identifying characteristics in few more slides to confirm the particular organized crude drug. Compare these characteristics with characteristics of the same powdered crude drug mentioned in the reference book.

PHYSICOCHEMICAL PARAMATERS-

Chromatographic fingerprint[5]-Separation, identification, impurity detection and assay of herbal drug in the formulation or in the extract are carried out by following methods: -HPTLC, HPLC/Densitometric chromatography, GLC, TLC

Importance-The herbal drug shows variability in its chemical constituents according to various locations/weather. To avoid any erroneous identification chromatographic fingerprint remains the assessment of choice.

Ash values[6]-The types of ash determined are Total ash, Acid insoluble and water soluble. Ash value is used to determine the quality and purity of the drug and to establish its identity. Ash contains inorganic radicals lie phosphates, carbonates, and silicates of sodium, potassium, magnesium, calcium, etc. These are present in definite amount in a particular crude drug, hence quantitative determination in terms of various ash values helps in their standardization. Ash value is used to determine foreign inorganic matter present as impurity.

Total Ash Value-The method of total ash is designed to determine the amount of material that remains after ignition. Ash is classified as physiological ash which is derived from the plant tissue itself and non-physiological ash which is the residue after ignition of extraneous matter (e.g. sand and soil).It is carried out at low temperatures possibly because alkali chlorides, which are

volatile at low temperatures, may be lost. The total ash consists of carbonates, phosphates, silicates and silica.

Acid insoluble ash- Sometimes, inorganic variables like calcium oxalate, silica, and carbonate content of the crude drug affects 'Total cash value'. Such variables are removed by treating with acid (as they are soluble in hydrochloric acid) and acid insoluble ash value is determined. Acid insoluble Ash, Water soluble ash and sulphated ash are also evaluated.

Extractive values[7]- It is useful for evaluation of a crude drug. It gives an idea about the nature of the chemical constituents present in a crude drug. Useful for estimation of constituents extracted with the solvent used for extraction. Employed for material for which as yet no suitable chemical or biological assay exists. It can be done by following methods: Cold maceration, hot extraction and ethanol.

Moisture content and volatile matter[8]- The moisture content of the drug should be minimized in order to prevent decomposition of crude drug either due to chemical change or microbial contamination.

The moisture content is determined by heating a drug at 105˚c in an oven to a constant weight. E.g. – Aloe should have moisture content not more than 10% w/w Moisture content can be determined by following methods- Gravimetric, Volumetric and instrumental.

Gravimetric method-Loss on Drying,

Volumetric-Azeotropic Toulene distillation method,

Instrumental- GC, NMR etc.,

Volatile oil content- Volatile oils are the liquid components of the plant cells, immiscible with water, volatile at ordinary temperature and can be steam distilled at ordinary pressure. Many herbal drugs contain volatile oil which is used as flavoring agent. For the drugs containing volatile constituents, toluene distillation method/steam distillation method is used to determine the volatile oil contents.

PHARMACOLOGICAL PARAMATERS[9]**- Bitterness value**- Medicinal plants having strong bitter taste are therapeutically used as appetizing agents .The bitterness is determined by comparing the threshold bitter concentration of an extract material with that of quinine hydrochloride .The bitterness value is expressed as unit's equivalent to the bitterness of a solution containing 1gm of quinine hydrochloride in 2000ml.

Method for determination-0.1gm of quinine hydrochloride is dissolved in 100ml drinking water and the stock solution is prepared. Then it is diluted and tested and compared with drug.

Bitterness value in unit per gm = 2000×C÷ A×B

Where, A = concentration of stock solution B = volume of test solution in tube with threshold bitter concentration C = quantity of quinine hydrochloride in the tube with threshold bitter concentration

Haemolytic property- Many medicinal plant materials, of the families Caryophyllaceae, Araliaceae, Sapindaceae, Primulaceae, and Dioscoreaceae contain saponins. The most characteristic property of saponins is their ability to cause haemolysis; when added to a suspension of blood, saponins produce changes in erythrocyte membranes, causing haemoglobin to diffuse into the surrounding medium. The haemolytic activity of plant materials, or a preparation containing saponins, is determined by comparison with that of a reference material, saponin R,

which has a haemolytic activity of 1000 units per g.

Determination- Calculate the haemolytic activity of the medicinal plant material using the following formula: 1000 ×a/b

Where, 1000 = the defined haemolytic activity of saponin R in relation to ox blood, a = quantity of saponin R that produces total haemolysis (g) b = quantity of plant material that produces total haemolysis (g)

Astringent property- It is determined by amount of tannins present in the drug .Tannins (or tanning substances) are substances capable of turning animal hides into leather by binding proteins to form water-insoluble substances that are resistant to proteolytic enzymes. This process, when applied to living tissue, is known as an "astringent" action and is the reason for the therapeutic application of tannins. Chemically, tannins are complex substances; usually occur as mixtures of polyphenols that are difficult to separate and crystallize.

Determination of Tannins[10] -Calculate the quantity of tannins as a percentage using the following formula: where w = the weight of the plant material in grams T_1=Weight of material extracted in water T_2=Weight of material not bound to hide powder T_0=Weight of hide powder material soluble in water that bind to standard frieberg Hide powder. $[T_1-(T_2-T_0)]\times 500/w$

Swelling Index- The swelling index is the volume in ml taken up by the swelling of 1 g of plant material under specified conditions. Its determination is based on the addition of water or a swelling agent as specified in the test procedure for each individual plant material (either whole, cut or pulverized). It gives an idea about the mucilage content of the drug; hence it is useful in the evaluation of crude drugs containing mucilage.

Foaming index- Many medicinal plant materials contain saponins that can cause persistent foam when an aqueous decoction is shaken. The foaming ability of an aqueous decoction of plant materials and their extracts is measured in terms of a foaming index. Calculate the foaming index using the following formula 1000/a : where a = the volume in ml of the decoction used for preparing the dilution in the tube where foaming to a height of 1 cm is observed. Saponins give persistent foam when shaken with water. Hence, plant material/extract containing saponins is evaluated by measuring the foaming ability in terms of foaming index.

Toxicological Parameters[11]-

Arsenic and heavy metals- Contamination of medicinal plant materials with arsenic and heavy metals can be attributed to many causes including environmental pollution and traces of pesticides. There are different methods to identify the amount and concentration of heavy metals in herbal drugs.

Limit test for arsenic and Limit test for cadmium and lead are few of them. The contents of lead and cadmium may be determined by inverse voltametry or by atomic emission spectrophotometry.

Determination-The following maximum amounts in dried plant materials, which are based on the ADI values, are proposed: ▫ lead, 10 mg/kg; ▫ cadmium, 0.3 mg/kg. Stain produced on $HgBr_2$ paper in comparison to standard stain.

Pesticide residues[12]-Examples of pesticide residues- Chlorinated hydrocarbons and related pesticides: BHC, DDT ,Chlorinated phenoxyalkanoic acid herbicides: 2,4-D; 2,4,5-T ,Organophosphorus pesticides: malathion, methyl parathion, parathion, Carbamate insecticides: carbaryl (carbaril), Dithiocarbamate fungicides: ferbam, maneb,

nabam, thiram, zineb ,Inorganic pesticides: calcium arsenate, lead arsenate ,Miscellaneous: ethylene dibromide, ethylene oxide, methyl bromide, Pesticides of plant origin: tobacco leaf and nicotine; pyrethrum flower, pyrethrum extract and pyrethroids; derris root and rotenoids. Includes total organic chloride and total organic phosphorous. Determination of pesticides- Pesticides should not be more than 1%, an ARL (in mg of pesticide per kg of plant material) can be calculated on the basis of the maximum acceptable daily intake of the pesticide for humans (ADI), as, recommended by WHO, and the mean daily intake (MDI) of the medicinal plant material. ARL=ADI×E×60/MDI×100 where ADI = maximum acceptable daily intake of pesticide (mg/kg of body weight); E = extraction factor, which determines the transition rate of the pesticide from the plant material into the dosage form; MDI = mean daily intake of medicinal plant product.

Microbial contamination- Contamination either at source or during processing is possible. Maximum possible limits of each organism are given in various texts. WHO limit for number of micro-organisms per gram of material

Type of micro-organ ism	Finished product	Raw material
E.coli	10^1	10^4
Salmonella	-	-
Total aerobic bacteria	10^3	-
Enterobacteria	10^3	-

Alfatoxins- Aflatoxins are naturally occurringmycotoxins produced mainly by Aspergillusflavus and Aspergillusparasiticus. The presence of aflatoxins can be determined by chromatographic methods using standard aflatoxins B1, B2, G1, G2 mixtures. Determination- IP method: NMT 2 µg/kg of aflatoxins B1& Total aflatoxins 4 µg/kg .USP method: NMT 5ppb of aflatoxins B1& Total aflatoxins 20ppb.

Radioactive contamination-The range of radionuclides that may be released into the environment as the result of a nuclear accident might include long-lived and short-lived fission products, actinides, and activation products. Microbial growth in herbals is usually avoided by irradiation. This process may sterilize the plant material but the radioactivity hazard should be taken into account. The nature and the intensity of radionuclides released may differ markedly and depend on the source (reactor, reprocessing plant, fuel fabrication plant, isotope production unit, etc.). The radioactivity of the plant samples should be checked accordingly to the guidelines of International Atomic Energy Agency (IAEA) in Vienna, Australia.

DISSCUSSION

Herbal drug standardization is very important for the safety and efficacy of the drug. The safety part can be compensated by performing various analysis on the drug, but the efficacy should not be judged only by the presence of the chemical compounds. There is a very fine line between efficacy and presence of compounds. The need of the hour is that Clinical/Laboratory analysis should be done to establish a relation between both of them. Herbal drug standardization should be done through multiple modes as the concentration of the phytochemicals varies according to climate, soil and environment. The database for the same should be maintained throughout the year and the data should be analyzed statistically for proper understanding of the medicinal plant. Newer aids of research should be used to identify minute variations. Finer analyti-

cal methods are now available which can be incorporated to analyze the herbal drugs. Gel electrophoresis of isolated and purified DNA samples is done to identify the herbal drug as it cannot vary with factors like climate, etc. DNA fingerprinting can be made the choice of analysis for a perfect assessment of authentification. Flow cytometry can also be applied for herbal drug identification, drug discovery and development.

CONCLUSION

The WHO guidelines are followed all over the world but the need of the hour is to update these principles with application of newer methods of analysis. The herbal drug assessment in Ayurveda is about the whole drug rather than concentrating on the active principles or phytoconstituents, thus finer methods of standardization should be developed. As Ayurvedic drugs are also included in the Drugs and Cosmetics Act, 1940 the drugs have to be safe and effective at the same time. This brings about the need for finer standardization of herbal drugs. We can certainly avoid the external contamination due to the pesticides by organic farming, heavy metal contamination by performing soil analysis and other tests, radioactive contamination is not very common in India but it can be prevented by using healthier ways of prevention.

REFERENCES

1. K R Khandelwal, 2013, PracticalPharmacognosy, 2nded, Nirali Prakashan, Pune,23.1-23.4.
2. SS Agarwal,M Paridhavi,2007. Herbal Drug Technology,FirstEd,Universities Press(India) Pvt. Ltd,Hybd.Pg629-630.
3. WHO Quality Control Methods for Medicinal Plant Materials, World Health Organization, Geneva, 1998
4. Kokate C.K., Gokhale, S.B. 2001. Practical Pharmacognosy. 2nd ed. NiraliPrakashan, Pune, p. 14-19. 2
5. Yvan Vander Heyden, Sept 1 2008,Extracting Information fromChromatographic HerbalFingerprints,LCGC Europe,Volume 21, Issue 9
6. Quality control of herbal drugs. 4th ed. Business Horizons, New Delhi, P. 184-219.
7. Ansari S.H. 2006. Essentials of Pharmacognosy. 1st ed. Birla Publications, New Delhi, p. 581- 596.
8. http://www.ncbi.nlm.nih.gov/pubmed/18396809.
9. Anonymous, 2010. Indian Pharmacopoeia. Vol.-3, Government of India, Ministry of Health and Family Welfare, New Delhi p. 2467-2472
10. Mukherjee P K & Peter J Houghton. 2009. Evaluation of Herbal Medicinal Products Perspectives on quality, safety and efficacy. Pharmaceutical Press London P, 19-23
11. Iqbal Ahmad, FarrukhAqil, and Mohammad Owais. Modern Phytomedicine, Turning Medicinal Plants into Drugs WILEY-VCH Verlag GmbH & Co. KGaA,Weinheim fGermany 2006 p.26-53
12. http://testinglaboratoryindia.com/ha-analytical-specifications.htm

CORRESPONDING AUTHOR

Dr Nilakshi Pradhan

Email: sumedhayk@yahoo.com

Source of support: Nil

Conflict of interest: None Declared