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A REVIEW: QUALITY CONTROL AND STANDARDIZATION OF HERBAL DRUG

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Abstract: The "herbal drug" is termed as a plant or part of plants that have been converted into phytopharmaceuticals by simply means of processes involving collection or harvesting, drying and storage. There is a great importance of herbal medicines in today's era. To avoid the adulteration and substitution standardization of herbal takes place. Standardization confirmed the quality and purity of herbal drugs. There are different analytical techniques for standardization of herbal drug like HPLC, HPTLC, TLC, GC-MS, LC-MS etc. Quality refers to the condition of a drug and is based on its collection, storage and processing. Definitions of three important pharmacopoeias such as identity, purity and active ingredient content. WHO provides guideline for herbal drug assessment 1) Quality control assessment 2) safety assessment 3) Toxicity assessment 4) Stability assessment. There are different evaluation parameters of herbal drug A) Physical Evaluation B) Chemical Evaluation C) Morphological Evaluation D) Analytical Evaluation etc. Different herbal medicine preparation available in market like Zandu Balm, Chyawanprash, Herbal shampoo, skin care products, Ointment, Oil, Liquid extract etc. Today around 80% population depends on herbal medicine. "The Considerable thing about herbal drug is that its Treatment always yield harmless benefits, not harmful effects."

Keywords: Standardization, Herbal drug, WHO, Evaluation method , Quality control.

I.INTRODUCTION

Traditional flavouring medication (HM) and its formulations are wide utilized in several fields for thousands of years. However, oriental countries like China, Republic of Korea and Japan square measure one in all the characteristics of oriental Chinese herbs. The preparation is that each one flavouring medicines square measure either one herb or a fancy flavouring assortment. The formula is extracted with boiling water throughout the boiling. this can be the most reason for internal control Oriental herbs square measure harder than western herbs medication. Like "General pointers for Methodology" For the study and analysis of ancient medication (World Health Organization, 2000) [27](Liang et al., 2004) the method of evaluating Quality, Purity of crude medication supported numerous factors like Morphological, microscopic, physical, chemical, Biological observation is termed as "Standardization". [4](Chopra & Sindhu, 2020) Standardization is that the adaptation of flavouring medication preparations. By adding excipients or by mix flavouring medicines or flavouring medicines, to an outlined level of a gaggle of ingredients or substances with well-known therapeutic effects. ready as a result of the plant extract is formed directly from the stuff. There square measure vital variations in composition, quality and therapeutic result. The standardized extract could be a consistent level of high-quality extract bound compounds and that they square measure subject to strict internal control throughout Growth, harvest and producing method stages.[7](Alamgir, 2017). internal control of herbals is of bigger importance for preservation of quality of the natural herbs and product. once the standard management side has identification of substance, adulterants, and substitutes; purity of material; and assay of active chemical constituent of bigger importance of the actual herb, then they're known as as pharmacopeial aspects of internal control.[2](Balekundri & Mannur, 2020). A drug is outlined as safe if it poses no well-known or potential hurt to the user. There square measure 3 security classes that ought to be thought-about to work out the sort of security necessities to be assured.

The 3 classes of are:

1. Category 1: long safety
2. Category 2: Safe beneath such that conditions of use (such flavouring medicines ought to otherwise be coated by well-established documentation)
3. Category 3: flavouring healthful product of unsure safety (The safety knowledge needed for this category of healthful product square measure similar to those for brand new substances).[10](Dhiman et al., 2016)

Need of Standardization :

1. The need for standardization of herbal medicine is important to achieve the potential quality and stability of the product.
2. If the quality and stability of the product is assured, the best quality of active ingredient has been used. But, the principle of standardization is not stated in the pharmacopoeia
3. Absence of quality standards has admitted an adverse effect which may lead to death.
4. To evaluate the parameters for standardization, specific devices are required as per GMP acquisition. Several factors like bio efficacy, reproducible therapeutic effects influence the standardization of herbal medicine.
5. The main factor is adulteration of herbal ingredients which can be done intentionally or unintentionally such as lack of storage, mixing of one ingredient with another, same name of herb or substitution with excipient material.
6. In the manufacture of allopathy products, herbal extracts are used as excipients, standardization is an essential procedure to be followed to test their bioactivity [4] (Chopra and Sindhu, 2020)

- **Advantages of herbal medicine:**
  - Low/minimal cost
  - Potency and efficiency
  - Increased resistance
  - More protection
  - Fewer side effects
  - Full accessibility
  - Recyclable

- **Disadvantages of herbal medicine :**
  - Cannot cure Quick Sickness and Accidents
  - Risks of self-medication
  - Complexity of standardization[23] (Maiti et al., 2011)

Drug Adulteration :

"Adulteration is that the observe of commutation the initial crude drug part or absolutely with another artificial substance however the latter is destitute of or inferior in chemical and therapeutic properties".

- **Methods of drug mixing:**

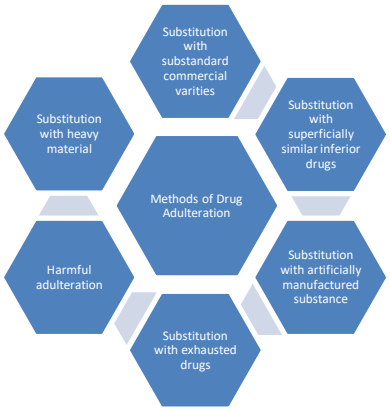


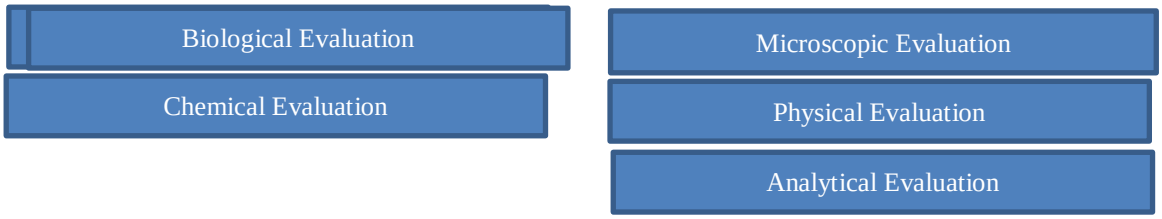
fig.1: methods of drug mixing

- **WHO Guideline for Standardization of Herbal Medicine:**

fig.2: who guideline for the evaluation of herbal drug.

- **Chemometrics:** A Swedish scientists Svante Wold was coined the term “Kemometrics” in Swedish and in English it is termed as “Chemometrics”. This termed was discovered in 1971. Chromatography is the most commonly used technique in standardization of herbal drugs. It is used in analysis of spectroscopic data and chemical information . The Principle Component Analysis(PCA) and Chemometric Resolution Method are the most commonly used techniques.[11](Khan & HN, 2016)

➢ **Methods of Drug Evaluation:**



1) **Organoleptic Evaluation/Morphological Evaluation:**

Organoleptic evaluation of drug by means of organ of sense (e.g. skin, eye, tongue, nose, ear.) or microscopic evaluation which include evaluation of drug by colour, odour, taste, size, shape and special features like touch, texture, etc . It is the technique of qualitative analysis based on the study of morphological and sensory profile of whole drug.

e.g. Aromatic odour of umbelliferous fruit, Sweet odour of liquorice, Fractured surface on cinchona , cascara bark.[13](R et al., 2016)

2) **Microscopic evaluation:**

It involves detailed examination of drugs and can be used to identify organized drugs by their known histological characters. It is mostly used for qualitative evaluation of crude drugs organized in absolute and potency forms with the help of microscopic. Various cellular tissues, trichomes, stomata, starch granules, calcium oxalate crystals and aleurone grains using microscope are some of the important parameters that play an important role in the identification of specific crude drugs[13](R et al., 2016).

Significance - This method allows detailed drug studies and is a tool for standard drug identification. is considered an important factor in the qualitative evaluation of organized herbal medicines.[8] (Prakash and Sayali, 2017).

a) **Palisade Ratio:** It is defined as average no. of palisade cells beneath each epidermal cell.

table 1: palisade ratio

Sr.no.	Name of Drug	Palisade ratio
1	Atropa belladona	05 - 70
2	Digitalis Lanata	2.5 – 6.5

b) **Stomatal number:** It is average number of stomata per square mm of the epidermis of the leaf

table 2: stomatal number

Sr no	Name of Drug	Stomatal no
1	Cotton	6
2	Ground nut	9
3	Tomato	7

C) Stomatal index : The Stomatal index is the percentage of the number of stomata formed by the total number of epidermal cells each stoma being counted as one cell.

Stomatal index = S/E+S×100

- Where S= total range of stomata in an exceedingly given space of leaf

E= range of cuticular cells within the same space of the leaf.

Table 3: Stomatal index

Sr no.	Name of Drug	Stomatal index
1	Mung bean	29.4
2	Sorghum	26.5

d) Vein islet number: The vein-islet number is average number of veinislet per square mm of leaf surface midway between midrib and margin. Various species of drugs are distinguished by vein-islet number E.g. The tinnevelly senna and therefore the senna are distinguished by the distinction within the range of veins, twenty seven and twenty two severally.

e) Vein termination number: It is defined as the no. of veinlet termination per sq. mm of the leaf surface midway between midrib and margin. .[8](Prakash & Sayali, 2017)

3)Physical Evaluation:

Physical constants are sometimes considered to evaluate a particular drug. These include water content, specific gravity, optical rotation, refractive power, melting point, viscosity, and solubility in various solvents. In addition, inclusions, total ash, acid-insoluble ash, water-soluble ash, swelling index, foam index, sequential extraction value, moisture content, viscosity, pH, disintegration time, friability, hardness, flowability, flocculation and sedimentation are also included. and alcohol content. All of these physical properties are useful in identifying and detecting constituents present in plants [13](R et al., 2016)

a)Ash value: The residue left after burning herbal remedies is known as the ash value, which represents naturally occurring minerals. Used to assess the purity of raw materials. It can be analyzed by determining various ash values. In herbal medicine, higher ash represents impurities.[4](Chopra & Sindhu, 2020)

Method: 2-3 g of ground drug is burned in a tared silica dish at a temperature not exceeding 450 °C, cooled and weighed. Calculate the ash content based on the air-dried drug. [8] (Prakash & Sayali, 2017)

table 4: example of ash value

Sr.no.	Name of drug	Total ash (%w/w)
1	Acacia Catechu	Not More Than 15%
2	Rawulfia Surpentina	Not More Than 8%

b) **Extractive Value:** This method is used to determine the amount of chemical constituents of crude drugs extracted with different solvents such as water-soluble extract, alcohol-soluble extract, ether-soluble extract.[4](Chopra & Sindhu, 2020)

table 5: examples of extractive value

Sr.no.	Name of drug	Alcohol soluble extractive value(%W/W)
1	Amla	Not More Than 40
2	Ashoka	Not More Than 15
3	Curcuma longa	Not More Than 8

c) **Moisture Content:** The amount of water contained in herbal medicines. It should be reduced to prevent drug degradation and estimation of the actual weight of the drug substance. The measurement method can be achieved by weighing an empty tarred porcelain bowl and pouring the appropriate amount of medicine into it. Place the porcelain mold in a convection oven at 105 °C for 5 hours until it reaches a constant weight [4] (Chopra & Sindhu, 2020).

table 6: example of moisture content

Sr. no.	Drug	Moisture Content%	W/W
1	Ajwain	Not More Than	10
2	Ashwaghandha	Not More Than	12
4	Sunthi	Not More Than	12

d) Refractive index : Refractive index gives an idea of purity. A ray of light bends as it travels from a thinner medium to a denser medium. This bending of light is called refraction. Therefore, the ratio of the speed of light in a vacuum to the speed of light in a substance is known as the refractive index of the second medium. It is considered an important tool for standardization as it is constant for liquids of a certain purity level. It is affected by the wavelength of incident light, temperature, and pressure. **[8]**(Prakash & Sayali, 2017)

table 7: example of refractive index.

Sr.no.	Drug	Refractive index
1	Caraway Oil	1.4838 – 1.4858
2	Clove oil	1.527 – 1.535

e) Determination of specific optical rotation : It depends on a phenomenon called polarization. Polarization means that light rotates clockwise, called right-handed, and counterclockwise, called left-handed, when the plane of polarization passes through the liquid. [8] (Prakash and Sayali, 2017)

It can be calculated using the formula:

$$D25 = 100 \times \varphi lc$$

Where, φ = observed rotation in drug at -25°

D = D line of sodium light

l = length of polarimeter tube.

c = concentration of substance in % w/v.

table 8: Specific optical rotation

Sr.no.	Drug	Angle Optical rotation
1	Caraway oil	+75 - +85
2	Clove oil	0 - +6
3	Honey	+3 - -15

f) Melting Point: Phytochemicals and herbal medicines have different melting points. It is relatively constant for phytochemicals and contains mixed chemicals for herbal medicines. [4] (Chopra & Sindhu, 2020)

Table 9: Example of Melting point

Sr.no.	Drug	Melting point
1	Colophony	75-85
2	Cocca butter	30-33
3	Bees wax	62-65

4) Biological evaluation: The pharmacological activity of specific drugs has been used to evaluate and standardize them. Assays on live animals and on intact or isolated organs can demonstrate efficacy of drugs or their formulations. These assays are known as biological assays or bioassays.[12] (M S Deepa 2016)

a) Determination of Bitterness Value:

Bitter medicinal plants are usually used therapeutically as appetite stimulants. Their bitter taste stimulates the gastrointestinal tract, especially the secretion of gastric juice. Bitter substances can be judged by taste. However, since most consist of two or more components with different bitterness, the total bitterness must first be measured by taste. The bitterness profile of a plant material is determined by comparing the threshold bitterness concentration of an extract of the material to that of a dilute solution of quinine hydrochloride. The bitterness value is expressed in units corresponding to the bitterness of a solution containing 1g of quinine hydrochloride in

2000ml. Safe drinking water should be used as a medium for extracting botanicals and to rinse the mouth after each tasting. Using distilled water will dull your taste buds quickly. Water hardness rarely has a significant effect on bitterness.[9](Kumar Bijauliya et al., 2017)

b) Determination of swelling index:

The swelling index is the amount (mL) absorbed by swelling of 1 gram of plant material under certain conditions. That determination is based on the addition of water or swelling agents specified in the individual plant material test specifications. After repeating the shaking for 1 hour using a graduated cylinder with a glass stopper, it is allowed to stand for the required time. Then the volume of the mixture is read. It is easy to mix the whole grass ingredients with the bulking agent, but the chopped or powdered ingredients must be shaken vigorously at regular intervals to evenly distribute the ingredients in the bulking agent.[9](Kumar Bijoulia et al., 2017)

c) Determination of foaming index:

Many medicinal plant materials contain saponins, and stirring an aqueous decoction can cause a persistent foam. The foaming power of aqueous decoctions of plant materials and their extracts is measured as foam index. [9](Kumar Bijoulia et al., 2017)

d) Determination of Pesticide Residues:

Pesticide residues are specific substances in food, agricultural products, or animal feed that result from the use of pesticides. Herbal medicines tend to contain pesticide residues resulting from agricultural practices such as spraying, soil behavior during cultivation, and addition of fumigant during storage. Various methods are used for the determination of pesticides by GC, MS, or GCMS. Some simple methods have also been published by WHO and the European Pharmacopoeia generally have limit values for pesticide residues in medicines. .[9](Kumar Bijauliya et al., 2017)

5) Chemical Evaluation:[6]

Most pharmaceuticals contain specific chemical ingredients on which their pharmacological and biological activities depend. A qualitative chemical test to determine drug quality and purity. Identification, isolation, and purification of active chemical constituents rely on chemical evaluation methods. A preliminary survey of phytochemicals is also part of the chemical evaluation. Qualitative chemical tests for chemical evaluation of herbal medicines include saponification number and acid number[15](Mr.Shailesh et al.2015).

a) Detection test for alkaloid:[8] (Prakash & Sayali, 2017)

1) Dragaondroff reagent
1) Mayer's reagent
2) Wagner reagent
3) Hager's test

b) Detection test for glycoside : [8] (Prakash & Sayali, 2017)

1) Modified brontragger's test
2) Legal test
3) Froth test
4) Foam test

c) Detection test for carbohydrates:[8] (Prakash & Sayali, 2017)

1) Molish test
2) Benedict's test
3) Fehling's test

d) Detection of Tannis : [8] (Prakash & Sayali, 2017)

1)Gelatin test
2)Gold bitter skin test

6)Analytical Evaluation:

Pharmacopeial monographs are the most practical approach to quality control of herbal medicines, and many of them exist (EMEA, 2005; WHO, 1998a,b, 1996a, 1998a, 1981). If pharmacopoeial monographs are not available, analytical method development and validation should be performed by the manufacturer. The best strategy is to strictly follow the pharmacopoeial definitions of identity, purity and potency or assays. Critical to meeting all monograph criteria is the need for appropriate analytical methods to determine identity, quality, and relative potency. A wealth of analytical methods is available there. However, among the analytical tools known to standardize monographs, chromatography is important, although it is often difficult to determine which one is the most suitablec.There are different analytical technique for the standardization of herbal drug like HPLC , HPTLC , TLC , GC , LC-MS, IR,Thermal analysis, GC-MS.[20](Kunle 2012).

a) High Performance Liquid Chromatography (HPLC):

Preparative and analytical HPLC is widely used in the following fields: Pharmaceutical industry for the isolation and purification of plant compounds. There are basically two types of HPLC primers. low pressure HPLC (usually under 5 bar) and high pressure HPLC (pressure >20 bar) [17](Ahmed et al., 2014). The main goal is to isolate the plant and as with any analytical work, the goal is to obtain information about the sample. Preparative HPLC is closer to analytical HPLC than conventional PLC, with higher column efficiencies and faster solvent velocities so you can perform your most difficult separations faster.[21] (Arun Rashid et al 2012). High Performance Liquid Chromatography (HPLC), also known as High Pressure Liquid Chromatography, is essentially a form of column chromatography that involves packing small particles (3-50µm) in a stationary phase with small bores (2-5mm). .), one end of which is connected to a source of pressurized liquid eluent (mobile phase). The three forms of high performance liquid chromatography are ion exchange, partition, and adsorption. In the research field of liquid chromatography, many new techniques have been developed recently to achieve better separations. These include capillary micellar electrokinetic chromatography (MECC), high-speed countercurrent chromatography (HSCCC), low-pressure size exclusion chromatography (SEC), reversed-phase ion-pair HPLC (RPIPC-HPLC), and strong ion-exchange HPLC (SAX). Is included. -HPLC). They offer new opportunities to successfully isolate multiple specific extracts of multiple herbal drugs. On the other hand, the advantage of HPLC lies in its versatility for analyzing compounds contained in herbal medicines. [13] (R et al., 2016)

b) High Performance Thin Layer Liquid Chromatography (HPTLC):

High performance thin layer chromatography is an improved version of thin layer chromatography. High Performance Thin Layer Chromatography is planar chromatography in which the separation of sample components on high performance layers with detection and detection is performed using advanced workstations, increasing the efficiency of the separation process [1](Beressa et al., 2021) HPTLC is appropriate for qualitative, quantitative and micro-preparative natural process. The HPTLC technique is wide utilized in the pharmaceutical business within the method of development, identification and detection of impurities in flavourer product and aids within the management of pesticides, phytotoxin content and quality of herbs and health foods. according Associate in Nursing HPTLC technique for the detection of withaferrin A and beta-sitosterol-diglucoside in four formulations of ashwagandha. antecedently HPTLC was accustomed discover, monitor and quantify bacosides A and B in Bacopa monaria which formula[24] (Bhupinder Singh Sekhon et al 2011). it's wide according that multiple samples will be analyzed at the same time exploitation fewer mobile phases than HPLC. There are reports that mobile phases of pH scale or higher will be used. Used for HPTLC. Another advantage of HPTLC is that the recurrent identification (sampling) of chromatograms till they're identical or totally different. HPTLC has so been investigated for the synchronic use of multiple elements in multicomponent formulations. this system will be accustomed standardize totally different plant species and assess their stability. and consistency of their formulation from totally different manufacturers[9](Kumar Bijoulia et al., 2017).

Steps concerned in HPTLC analysis[19](Shivatare et al., 2013).

- choice of stationary section
- Selection of mobile section
- Sample preparation and application
- recording Development (separation)
- Investigation

c) Infrared Spectroscopy :

IR spectroscopy was used to determine the functional groups present in the samples. Infrared absorption spectroscopy is the measurement of the wavelength and intensity of light absorption in the near-infrared range of a sample. Mid-infrared light has enough energy to excite nuclear vibrations to higher energy levels. The wavelengths of some IR absorption bands are characteristic of certain types of chemical bonds, and IR spectroscopy has shown the greatest utility in the qualitative analysis of organic molecules and metals [1] (Beresa et al., 2021). The IR method is widely used in the herbal medicine industry as a quality control tool for direct identification and quantification of herbs or extracts. Different powders of Panax Notoginseng were identified using FT-IR and 2D FT-IR and the combination of ATR-FTIR microscopy was directly applied to determine the complex composition of herbal powders. [3] (BMMSHK et al., 2020)

d) Thin Layer Chromatography (TLC) :

TLC is a technique in which a solute is partitioned between two phases, a stationary phase acting by adsorption and a mobile phase in liquid form. The sorbent is a relatively thin, uniform layer of finely divided drug material applied to a glass, plastic, or metal foil/plate. glass plates are most commonly used. Separation can also be achieved based on a combination of partition/distribution and adsorption, depending on the particular support and its use with different solvents. It can be identified by observing spots on the sample plate with the same Rf value and size obtained for the unknown and reference samples. A visual comparison of spot size and intensity is usually used for semi-quantitative estimation. TLC has the advantage of having different ways to detect herbal medicines in the analysis. Also, TLC is fairly simple and can be used to analyze multiple samples. More than 30 sample spots can be interrogated for each plate. Useful qualitative and quantitative information can be obtained from the developed TLC plates using the CA MAG video storage system and the TLC-QA-UV method.[13](R et al., 2016).

e) Gas Chromatograhy – Mass Spectroscopy :

The Gas Chromatograph can be easily connected to various types of Rapid Scan Spectrometers. Capillary column flow rates are generally low, but sufficient for the column. The output can easily be fed directly into the MS ionization chamber. Among them, the simplest mass detector in GC is the ion trap detector. An efficient, rapid and accurate capillary gas chromatography method was used to determine organochlorine pesticide residues. SPE extracts were separated by capillary column using an electrochemical detector. The fractionation ratio was 1:2.2 using N 2 carrier gas at a flow rate of 1.4 mL/min. The injector temperature is 220 °C and the detector temperature is 330 °C. By comparison, the organochlorine pesticides gave good linearity. It has been used to identify numerous constituents present in natural and biological systems [21] (Arun Rasheed et al 2012). The advantages of GC-MS are: (1) With capillary columns, GC-MS generally has very good separation power and can produce high quality chemical fingerprints. (2) Combining mass spectrometry with a corresponding mass spectrometry database, the GC-MS provided relative qualitative and quantitative information about the composition of the studied herb, aiding further studies to elucidate relationships. Very helpful. Further research between the chemical constituents of herbal medicines and their pharmacology. Therefore, in our opinion, GC-MS should be the best tool for the analysis of volatile compounds in herbal medicines.[18](Farooqui et al., 2014)

f) Liquid Chromatography – Mass Spectroscopy:

LC-MS has become the method of choice at many stages of drug development. Recent advances include e-beam, thermal spray, and ion implantation, liquid secondary ion mass spectrometry, followed by laser mass spectrometry at 600 MHz, which offer the unique advantages of high detection sensitivity and specificity, allowing molecules to be Allows accurate identification of the proteins and peptides used. weights are enabled. Isotope patterns can be detected with this technique.[22] (Nikam et al., 2012)

g) DNA Fingerprint Technique :

DNA analysis has proven to be an important tool in the standardization of herbal medicines. This technique helps distinguish between phytochemically indistinguishable genuine drugs and alternatives or adulterations. DNA fingerprinting genomes have been reported to remain the same regardless of the plant part used, whereas phytochemical levels vary with the plant part used, physiology and environment.[22](Nikam et al., 2012)

h) Thermal Analysis of herbal drugs:

Differential scanning using thermogyitravimetric analysis (TGA), differential thermal analysis (DTA) and calorimetry (DSC), physical or chemical changes in various products including herbal and pre-formulation or pharmaceutical research. Compatibility of excipients. [26](Gordalla, 2010). Thermal analysis is a term encompassing a group of techniques used to monitor the physical or

chemical properties of a substance or its reaction products as a function of time or temperature, the temperature of the sample being controlled programming exposed to the atmosphere below. The use of thermal analysis techniques is increasing in the field of drug development, where they are used for excipient characterization and formulation development. For compatibility testing, this analysis allows to observe the thermal degradation profile of drugs, excipients, and their mixtures. Factors such as drug protection, reactions, intermediate and final degradation products generated, and degradation kinetics can be assessed.[5] (Dourado, 2019)

Omics: A New Technique in Herbal Drug Standardization and Quantification:

The Omics study contains a large number of scores for each endpoint, giving you comprehensive and integrated knowledge while viewing different factors separately. Omic technology is primarily used to identify biomedical resources such as: 1) Genomic technique in DNA sequencing 2) Fingerprinting or DNA microarrays. Omics technology is the genome, transcriptome (total number of genes converted into transcripts (i.e., mRNA molecules)), proteome (total proteins found in a given cell or tissue), metabolome (total metabolites and intermediate stages of a cell or tissue), interactome (position of molecules such as biologically active metabolites that interact with a particular protein), and phenomenon (the sum of all observable features of an organism) level[14](Pandey et al., 2016).

Conclusion:

Quality control of herbal medicines aims to ensure their quality, safety and efficacy[25](Kushwaha et al., 2010). Chemical makers are central to our current quality control operations. fluorescence quenching, Combining Chromatography and Spectrophotometry, Biological Assays, Use of Biomarkers in Fingerprinting, and More newer technique available for the standardization of herbaldrug.[16](Factor et al., 2014)

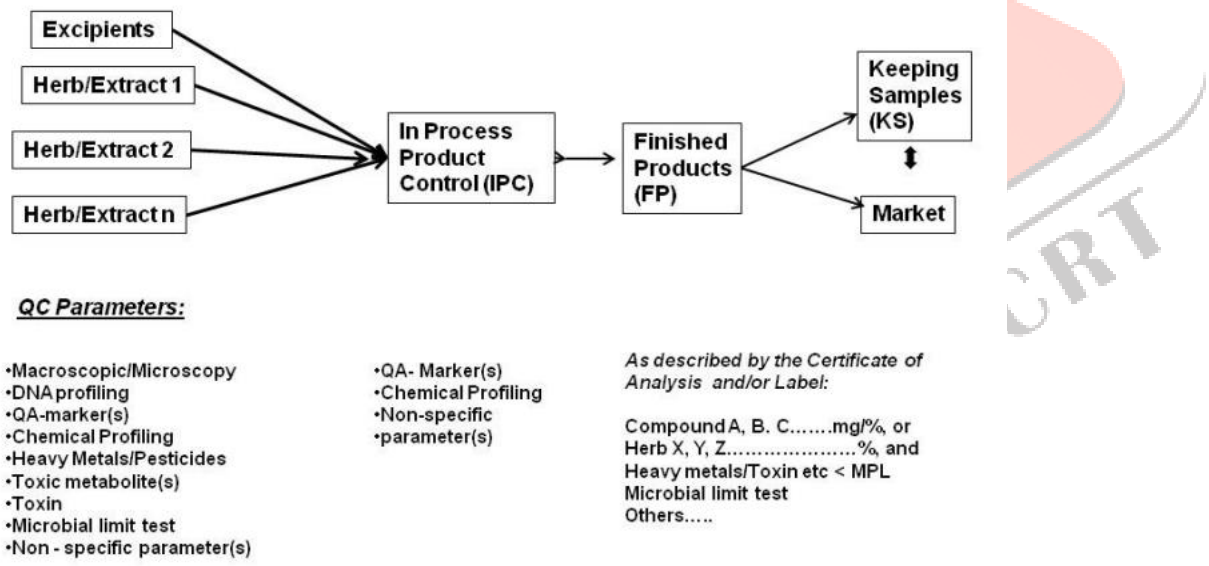


fig.3: complete qc parameters for every stage in the production line of herbal drugs[6](indrayanto, 2018).

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