A REVIEW ON DIFFERENT ANALYTICAL TECHNIQUES TO DETECT HEAVY METALS IN AYURVEDIC PREPARATION

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***Abstract*: Ayurvedic medicines contains heavy metals such as Lead, Cadmium, Mercury, Arsenic, Zinc, Copper. The human body require certain amount of trace element. If the amount of metals increases in human body it can cause toxic effect like cancer, lung disease, liver dysfunction etc. Thus, detection of heavy metals in Ayurvedic preparation is of great concern. In recent years many analytical techniques like Atomic absorbance spectrophotometry (AAS), Mass spectroscopy (MS), Inductive coupled plasma mass spectrometry (ICP-MS), Energy- dispersive X-Ray fluorescence spectroscopy (EDXRF), Gas chromatography (GC), Flame atomic absorption spectroscopy (FAAS), Inductive coupled plasma optical emission spectroscopy (ICP-OES) are the best techniques for detection of heavy metals in Ayurvedic preparation. This article will also focus on the principle of analytical method to reveal its importance.**

***Keywords:* Heavy metals, Ayurvedic preparation, Analytical Techniques, AAS, MS, ICP-MS, EDXRF, GC, FAAS, ICP-OES**

1. INTRODUCTION TO AYURVEDIC PREPARATION

Ayurveda is a holistic system of medicine that has been practiced for thousands of years. It promotes balance among body, mind, spirit, and the environment. People who practice Ayurveda often use herbal compounds (Ayurvedic medicines) to treat ailments and promote health.[1]

Ayurveda, also called Ayurvedic medicine, traditional system of Indian medicine. Ayurvedic medicine is an example of a well-organized system of traditional health care, both preventive and curative, that is widely practiced in parts of Asia.

Ayurveda has a long tradition behind it, having originated in India perhaps as much as 3,000 years ago.

Today it remains a favored form of health care in large parts of the Eastern world, especially in India, where a large percentage of the population uses this system exclusively or combined with modern medicine.[2]

Fig.1. Ayurvedic medicine

➢ **AYURVEDIC FORMULATION AVAILABLE IN MARKET**

- Tablet
- Churna
- Syrup
- Gel
- Powder
- Bhasma
- Yastimadhu
- Aloevera
- Haritaki
- Ahwagrasna

- Nirgundi
- Chanderprapha
- JasadBhasma
- Mentha
- Foenugreek
- Sweet basil
- Hibiscus
- Cumin
- Withaniasomnifera

2. INTRODUCTION TO HEAVY METALS

➢ DEFINITION

The term heavy metal refers to any metallic chemical element that has a relatively high density and is toxic or poisonous at low concentrations. Examples of heavy metals include mercury (Hg), cadmium (Cd), arsenic (As), chromium (Cr),and lead (Pb).[3]
The human body requires a number of trace elements like Calcium(Ca), Manganese(Mg), Aluminium(Al) etc. in order to maintain good health. These trace elements, essential for human nutrition. In human beings, these elements are mostly required in amounts less than 100 milligrams per day.
On the other side, several scientific report have indicated that herbal medicines also contain the toxic heavy metals which can cause various toxic effects like cancer, liver dysfunction, lung disease, cerebral hemorrhage, alopecia etc.
According to the WHO (World Health Organization, 1991), lead, cadmium, chromium, and other heavy metals must definitely be controlled in medicines in order to assure their safety. [4]

➢ TYPES

- Lead(Pb)
- Cadmium(Cd)
- Copper(Cu)
- Iron (Fe)
- Nickel(Ni)
- Mercury(Hg)
- Chromium(Cr)
- Meganese(Mn)
- Arsenic(As)
- Zinc (Zn)

➢ Limit of heavy metals in ayurvedic product as per WHO, 2005[5]

Table no 1. limit of heavy metals in ayurvedic as per WHO

COUNTRIES	HEAVY METALS				
	Arsenic (AS)	**Lead (Pb)**	**Cadmium (Cd)**	**Cromium (Cr)**	**Mercury (Hg)**
India	3 ppm	10 ppm	0.3 ppm		1 ppm
China	2 ppm	10 ppm	1 ppm	-	0.5ppm
Singapore	5 ppm	20 ppm			0.5 ppm
Thailand	4 ppm	10 ppm	0.3 ppm	-	-
Canada	5 ppm	10 ppm	0.3 ppm	2	0.2 Ppm

➢ DISADVANTAGES OF HEAVY METALS IN AYURVEDIC PREPARATION

Some Ayurvedic medicines and other traditional medicines may contain harmful heavy metals. one out of every five Ayurvedic medications purchased online contained lead, mercury, or arsenic. In recent years, several children of Indian decent in Minnesota have had high levels of lead or mercury in their blood that were traced back to Ayurvedic medicine.[1]

1. LEAD

Acute lead poisoning is very serious and is usually caused by a recent exposure to a high amount of lead. The symptoms may include

- stomach pains
- constipation
- tiredness
- nausea and vomiting
- headache
- seizures.

Extremely high blood lead levels can cause long-term organ damage and even death. A person who is exposed to smaller amounts of lead over a longer time period may have symptoms such as:

- irritability
- behavioural problems
- lack of energy
- raised blood pressure
- loss of appetite
- Poor coordination.[6]

2. ARSENIC

Ayurvedic medicines are known to contain arsenic and concentrations up to toxic levels have been reported in certain formulations. However, clinical disease due to arsenic containing ayurvedic medicines has rarely been reported. We seek to highlight the existence of toxic levels of arsenic in certain ayurvedic preparations that can produce serious systemic manifestations.[7]

Long-term exposure to arsenic can cause more severe symptoms. You should seek emergency help if you experience any of the following after a suspected arsenic exposure:

- Darkening skin
- Constant sore throat
- Persistent digestive issues.[8]

3. MERCURY

Exposure to metallic mercury vapors at higher levels for shorter periods of time can lead to

- diarrhea,
- nausea,
- skin rashes,
- increased heart rate or
- blood pressure.
- Vommiting
- Lung damage

Symptoms of organic mercury poisoning include

- depression,
- memory problems,
- tremors,
- fatigue,
- headache,
- hair loss, *etc.* [9]

4. CADMIUM

Cadmium is a heavy metal of considerable toxicity with destructive impact on most organ systems. Human exposure to Cd occurs chiefly through inhalation or ingestion. Ten to fifty percent of inhaled cadmium dust is absorbed, depending on particle size.

- Hypertension
- Diabetes
- Sudden cardiac death
- Myocardial infarction
- Pancreatic cancer [10]

5. ZINC

Compared to several other metal ions with similar chemical properties, zinc is relatively harmless. Only exposure to high doses has toxic effects, making acute zinc intoxication a rare event. [11]

- Nausea
- Vomiting
- Epigastric pain
- Lethargy
- Weakness. [12]

6. COPPER

Copper is a heavy metal that's perfectly safe to consume at low levels. You have about 50 to 80 milligrams (mg) of copper in your body that's mostly found in your muscles and liver, where excess copper is filtered out into waste products.

The normal range for copper levels in the blood is 70 to 140 micrograms per deciliter (mcg/dL).Your body needs copper for a number of processes and functions.

Copper helps develop tissues that make up your bones, joints, and ligaments. You can get plenty of copper from your diet.Copper toxicity means you have more than 140 mcg/dL of copper in your blood.

Copper poisoning may cause the following mental and behavioral symptoms:

- feeling anxious or irritable
- having trouble paying attention
- feeling overexcited or overwhelmed
- feeling unusually sad or depressed
- sudden changes in your mood

other symptoms like

- headaches
- fever
- passing out
- feeling sick
- throwing up
- blood in your vomit
- diarrhea
- black poop
- abdominal cramps
- brown ring-shaped markings in your eyes (Kayser-Fleischer rings)
- yellowing of eyes and skin (jaundice)

Long-term copper toxicity can also be fatal or cause:

- kidney conditions
- liver damage or failure
- heart failure
- brain damage
- Gastrointestinal and
- kidney dysfunction,
- nervous system disorders,
- skin lesions,
- vascular damage,
- immune system dysfunction,
- birth defects. [13]

3. ANALYTICAL METHOD USED FOR DETECTION OF HEAVY METALS IN AYURVEDIC PREPARATION

1) Atomic absorbance spectrophotometry (AAS)
2) Mass spectroscopy (MS)
3) Inductive coupled plasma mass spectrometry (ICP-MS)
4) Energy- dispersive X-Ray fluorescence spectroscopy (EDXRF)
5) Gas chromatography (GC)
6) Flame atomic absorption spectroscopy (FAAS)
7) Inductive coupled plasma optical emission spectroscopy (ICP-OES)

1) ATOMIC ABSORBANCE SPECTROPHOTOMETRY (AAS)

➢ **PRINCIPLE**

Atomic absorption spectroscopy (AAS) is a Spectro analytical procedure for the quantitative determination of chemical elements using the absorption of optical radiation (light) by free atoms in the gaseous state. Atomic absorption spectroscopy is based on absorption of light by free metallic ions.

In analytical chemistry the technique is used for determining the concentration of a particular element (the analyte) in a sample to be analyzed.

The technique makes use of the atomic absorption spectrum of a sample in order to assess the concentration of specific analytes within it. It requires standards with known analyte content to establish the relation between the measured absorbance and the analyte concentration and relies therefore on the Beer– Lambert law.[14]

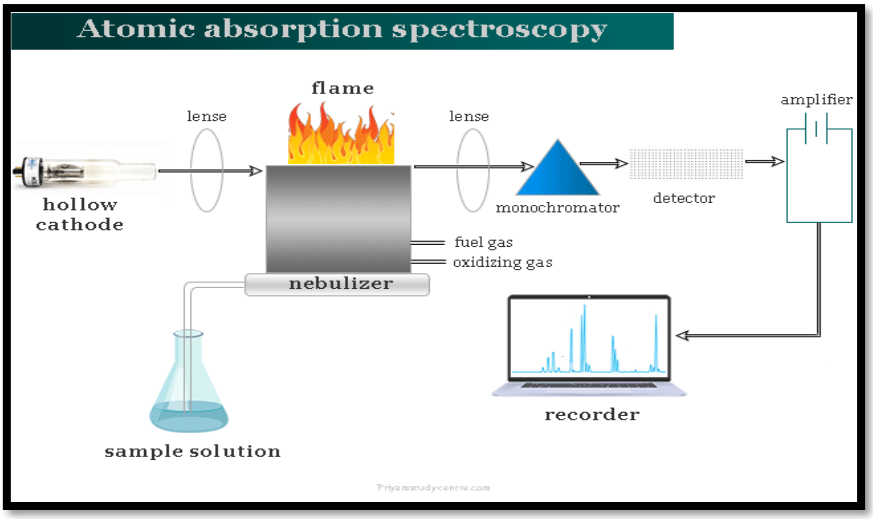

Fig .2. Schematic diagram of AAS instrument

2) MASS SPECTROSCOPY (MS)

➢ **PRINCIPLE**

Mass spectrometry (MS) is an analytical technique that separates ionized particles such as atoms, molecules, and clusters by using differences in the ratios of their charges to their respective masses (mass/charge; *m/z*), and can be used to determine the molecular weight of the particles. [15]

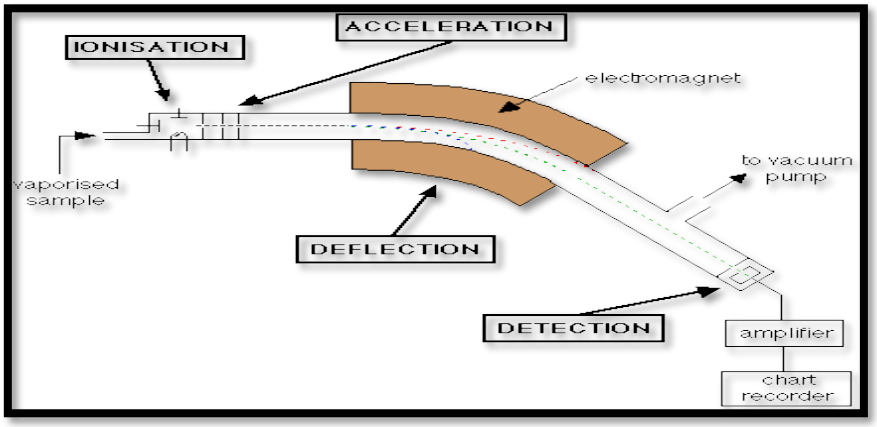

Fig.3. Schematic diagram of MS instrument

3) INDUCTIVE COUPLED PLASMA MASS SPECTROMETRY (ICP-MS)

➢ **PRINCIPLE**

Inductively coupled plasma mass spectrometry (ICP-MS) is a type of mass spectrometry that is highly sensitive. It can see a range of metals and several non-metals at concentrations below one part in 10^{12} (part per trillion). It is based on hooking together an inductively coupled plasma as a method of producing ions (ionization) with a mass spectrometer as a method of separating and detecting the ions. Many chemists use Argon as a carrier gas to make the plasma. The machine sends the sample ions through a series of small cones. The cones let the plasma ions slowly enter the vacuum chamber of the mass spectrometer.[16]

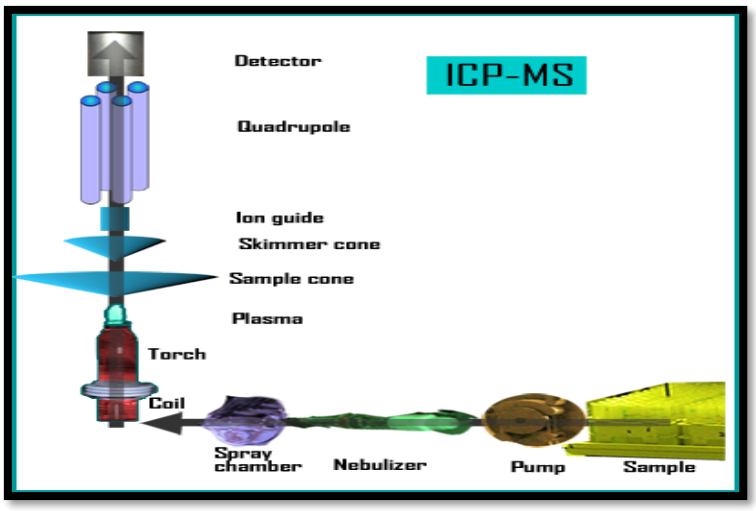

Fig.4. Schematic diagram of ICP-MS instrument.

4) GAS CHROMATOGRAPHY (GC)

➢ PRINCIPLE

Components in the mixture are distributed between two phases, one of which is a stationary phase, and the other is a mobile phase gas, or carrier gas, that carries the mixture through the stationary phase. Compounds in the mobile phase interact with the stationary phase as they pass through. Due to the differences in properties and structures of each component, the size and affinity of each interaction with the stationary phase are different. Therefore, under the same driving force, the retention time of different components differs in the column, thus moving out of the column in different orders.[17]

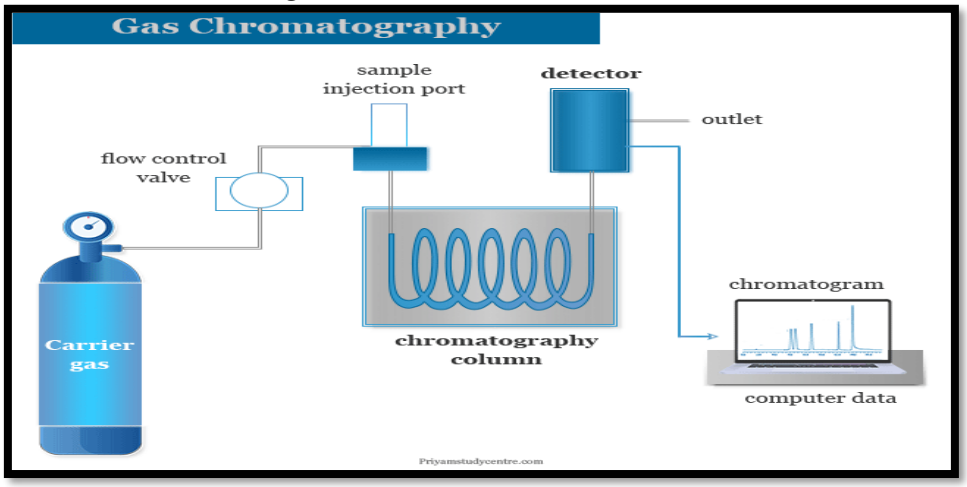

Fig.5. Schematic diagram of GC instrument.

5) ENERGY DISPERSIVE X-RAY FLUORESCENCE SPECTROSCOPY (EDXRF)

Energy-dispersive X-ray fluorescence (EDXRF) spectrometry exhibits several advantages over other methods that measure elemental content. This technique can measure numerous sample forms and can satisfy different measurement requirements . Moreover, EDXRF spectrometry can detect a wide range of elements, even several elements simultaneously. This technique is nondestructive, fast, highly accurate, and environment friendly.

EDXRF spectrometry can be used on different types of sample, such as bulk, liquid, powder, and gas.

➢ PRINCIPLE

Direct excitation is a process by which atoms in a specimen are excited by primary photons from external sources, such as an X-ray tube, radioactive source, and synchrotron beam, to produce primary fluorescence.[18]

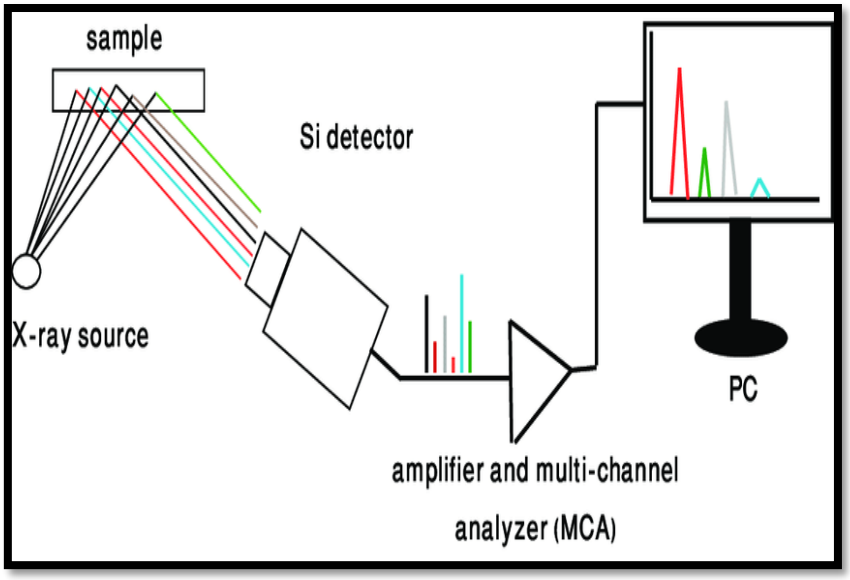

Fig.6. Schematic diagram of EDXRF instrument

6) FLAME ATOMIC ABSORPTION SPECTROSCOPY (FAAS)

➢ PRINCIPLE

When a solution of metallic species in sprayed into a flame, the fine droplet s of metallic species will be obtained. Due to thermal energy of the flame, the solvent in the droplets evaporate, leaving a fine residue, which are converted to neutral atoms.

These neutral atoms are converted into exited state atoms by thermal energy of the flame. As the exited state is not stable, these exited atoms returns into ground state, with the emission of radiation of specific wavelength.

The wavelength of the radiation emitted of radiation is characteristics of the element and is used to identify the element.[19]

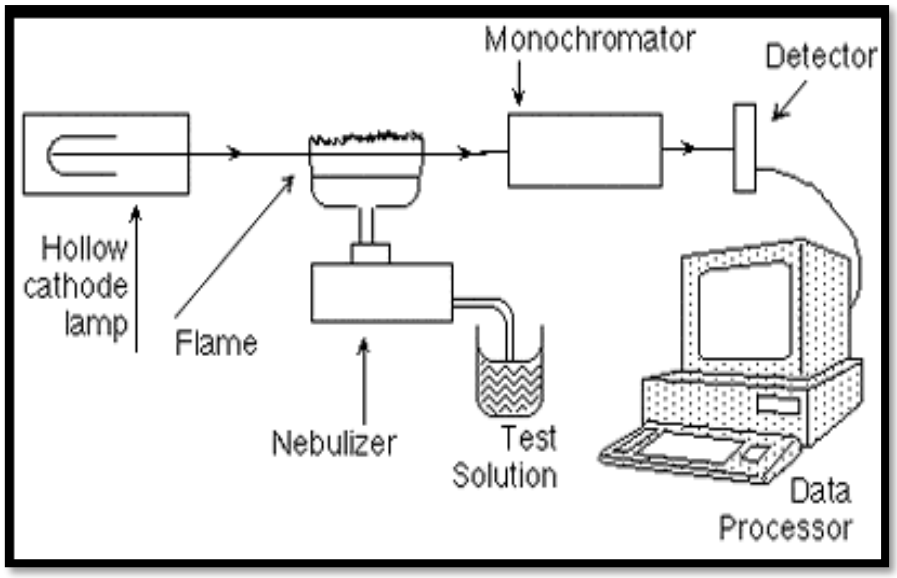

Fig.7. Schematic diagram of FAAS instrument

7) Inductive coupled plasma Optical emission spectroscopy (ICP-OES)

ICP-OES has been commercially available since 1974 and detects elements for sample by using plasma (the fourth state of matters, next to solid, liquid & gas) and spectrometer. This instrument consists of a light source, detector, spectrometer and data processing unit.

➢ **PRINCIPLE**

The basic principle is when plasma energy is provided to a sample from outside, the component elements are excited.
Emission rays are released when the excited atoms return to low energy position and the emission rays that correspond to the photon wavelength are determined by the spectrometer.
The element type is measured depending on the position of the photon rays, and the component of each element is determined based on the intensity of the rays.[20]

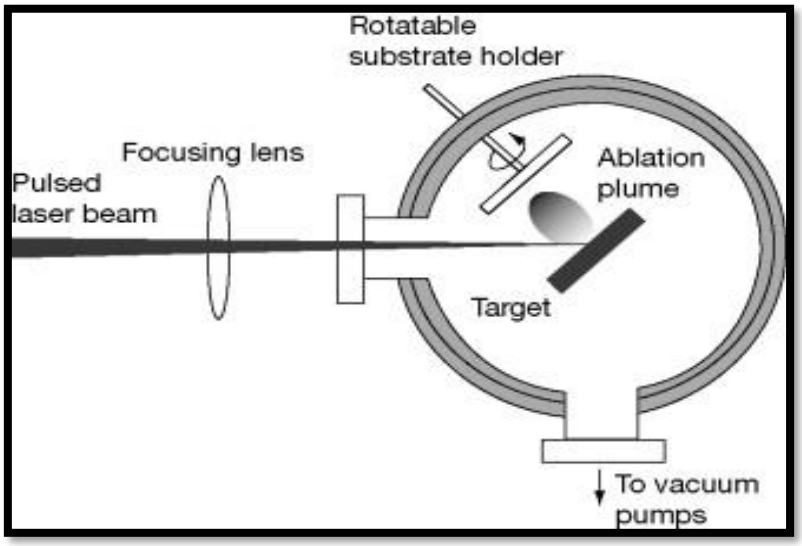

Fig.8 Schematic diagram of OES instrument[30]

4. REVIEW OF LITERATURE

1.Huang Q , chen S and yang C (2021) were developed **Inductively Coupled Plasma Optical Mass Spectrometry (ICP-MS)** to estimate the heavy metals such as Cd, Pb, As, Hg and Cu in herbal medicine**.** Pb, Cd, As, and Hg have resulted in higher than acceptable risks in 25 kinds of herbs. The maximal Estimated Daily Intake of Pb in seven herbs, of Cd in five, of Hg in four, and As in three exceeded their corresponding Provisional Tolerable Daily Intakes.[21]

2. Quds T and Ahmed M and Shakeel S and JalbaniN (2021) were developed **flame atomic absorption spectrometry (FAAS**) to detect heavy metal such as; arsenic (As), Cd, Pb and Hg, in different dosage forms of herbal medicines such **as** crude forms of syrups, gel, capsule, powder and tonic. The investigated results displayed the As level (0.00 ppm to 0.580 ppm); Cd (0.001 ppm to 0.006 ppm); Pb (0.00 ppm to 1.078 ppm) and Hg (0.001 ppm to 0.012 ppm). All results were found below the permissible limit of acceptability intake of the World Health Organization (WHO) and American Herbal Products Association (AHPA). In Conclusion the findings reveal that the investigated herbal products available in Pakistan are safe with reference to heavy metals, and considered non-toxic for human consumption.[22]

3. Harshita P, Rajeshwari M (2020) were developed **Atomic absorption spectroscopy (AAS)** for estimation of heavy metals such as Cd, Pb, As in marketed and in house formulations of udarkalp Churna. The result revealed that lead concentrations varied

from 0.15 to 0.165 ppm, Cd concentrations ranged from 0.05-0.07ppm and Arsenic concentrations were in between 0.25-0.34 ppm. According to WHO guidelines, the permissible limits for Pb, Cd and As are 10ppm, 0.3ppm and 10 ppm respectively. It can be concluded that the marketed and in house formulations of the UdarkalpChurna was in accordance with the standards laid down by the WHO in terms of heavy metals concentrations.[23]

4. **Abualhasan M, Jaradat N and Sawaftah Z and Mohsen H(2019)** were developed **Atomic absorption spectroscopy (AAS)** for estimation of heavy metals such as copper, Cd, Zn, Pb in Selectedherbals medicine such as (Chamomile, Ginger, Hawthorn, Roselle, etc). The Results showed That the result of the heavy metals testing showed that copper and cadmium were above the allowable limits in all the tested plants. the average value was 56.52 and 64.01 ppm respectively. lead was within the acceptable limits in all the tested plants (average =0.3210 ppm). Zn metal was above the allowable limit in 78.9% of the tested samples. In conclusion Herbal medicines used in the Palestinian markets don't meet international requirements. Urgent action has to be taken by the responsible authorities such as implementing importation and registration requirements. [24]

5.Daisy Joseph (2018)were developed **Energy Dispersive X-ray Fluorescence (EDXRF)** Technique for estimation of heavy metals such as Zinc in nine preparations of JasadBhasma along with zinc oxide. JasadBhasma preparations prepared using different processes showed significant levels of Zn (22% to 48% respectively. The other trace elements were also detected at significant level in these preparations. It is concluded that a uniform procedure need to be adopted to bring uniformity in the quality of Ayurvedic drugs.[25]

6. Vijayalakshmi S and kripa KG (2018) were developed **Inductively coupled plasma optical emission spectrometry (ICP-OES)** to estimate heavy metals such as Cd, Cr, Mg, Ni, and Pb and Hg, Fe, Zn, Cu, and Co in Blepharismaderaspatensiss (L.)Heyneex Roth commonly called as Nethiramooli in Tamil is a widely distributed medicinal herb. Results indicated that the toxic metal, Hg was not detectable even in traces; other metals such as cadmium, chromium, molybdenum, manganese, nickel, and lead were found at very low quantities while trace elements such as Fe, Zn, Cu, and Co in fairly significant concentrations. In Conclusionsthese results thus substantiate the safe usage of this plant and may be used to set new standards for prescribing the dosage of the herbal drugs prepared from it, ensuring a better understanding of the risks associated with its usage.[26]

7.Thomas A and Kumar D (2018) were developed **inductively coupled plasma MassSpectrometry (ICP-MS)**. In this,ayurvedic tablet were selected to the detection of heavy metals including; lead, arsenic, cadmium and mercury. The result showed that Among the 16 Ayurvedic tablets analyzed, 4 failed for Pb, 2 for As and 6 for Hg. All the tested samples had Cd below the limit set by Government of India. In conclusion tablets are the worst affected ayurvedic dosage form.[27]

8.Nwachukwu R, And Janefrances N and Jude I (2018) Were developed **Atomic absorption spectrophotometer (AAS)**using 25 popularly consumed types of commercially available herbal drugs in Southern Nigeriato detect heavy metals including; Fe, Cd, Zn, Ni, Co and Pb. The results showed that the provisional weekly intake (PWI) of the metals did not exceed the recommended provisional weekly tolerable intake (PTWI) except nickel in 5 samples. Seventeen brands of the studied herbal drugs had Fe, Cd, Ni, Cu and Pb levels higher than the WHO/FAO limits. [28]

9.Ismail I, Gezawa I, Nafisatu K and Abdulkadir I (2018) were developed **Chromatography-Mass Spectrometric (GC-MS) analytical** foe estimation heavy metals such as Co, Cr, Co, Fe, Mg, Ni and Zn, Cd, As, Hg, and Pb in Gasca D herbal formulation. Result of the present study showed no traces of Pb, Hg, As, and Zn. The level of Cd andCr and Mn, Nifound in the present study was much lower than the permissible limit. In conclusion this study the level of some toxic metals was measured and found the most commonly toxic metal to be below the detection limit or in trace amount.[29]

10. **Lee J , Kim S and Jung K , Ryu H (2017)** were developed **inductively coupled plasma mass spectrometry (ICP-MS), gas chromatography/nitrogen phosphorous detector (GC/NPD), gas chromatography/micro electron capture detector (GC/µECD)** method to determine heavy metals such as Cd, As, Hg, and Pb in traditional herbal decoctions. The results shows that One hundred fifty-two of One hundred fifty-five decoctions (98.1%) contained one of three heavy metals (96.1% for As, 97.4% for Cd, and 90.3% for Pb, 0.0% for Hg). In Conclusions This study support that the contained status of toxic heavy metals in herbal decoctions are currently within safe level in Korea. [30]

11. **Iqbal S, Gurumurthy P and Pillai K (2015)** were developed **Gas chromatography – mass spectroscopy and inductively coupled plasma atomic emission spectroscopy (ICP-AES)** to determine heavy metals such asAs, Cr, Pb in polyherbal formulation. The result showed that the PHF not contains high toxic level of heavy metals; (As -0.022 mg/L; below detectable level, Cr 0.026 mg/L and Pb 0.006 mg/L; below detectable level) and is safe for the administration into the body.[31]

12.Akram S, Najam R and Rizwani G (2015) were developed **Atomic Absorption Spectroscopy (AAS)** for determination of heavy metals such as Cu, Ni, Zn, Cd, Hg, Mn, Fe, Na, Ca, and Mg in the herbal plants like (H. Integrifolia, D. regia, R. communis, C. equisetifolia, N. oleander, T. populnea, M. elengi, H. schizopetalus, P. pterocarpum) and an antidiabetic Malaysian herbal drug product containing (Punicagranatum L. (Mast) Hook, Momordicacharantia L., Tamarindusindica L., Lawsoniainermis L.)**.**the result indicate that All the metals under study were within the prescribed limits except mercury.[32]

13.**Szok A, Kurzawa M, Szlyk E (2014**) were developed **inductively coupled plasma mass spectrometry (ICP-MS)** to estimation of heavy metals such as Cd, Pb, As, Ni in Traditional Chinese Medicine (Angelica sinensis, Bacopamonnieri, Bupleurumsinensis, Curcuma longa, Cola accuminata, Emblicaofficinalis, Garciniacambogia) and some European herbs (Echinacea purpurea, Hypericumperforatum, Vitisvinifera). Results indicate that, the highest concentration of Pb and Cd was found. In conclusion studied medicinal plants contain elementthat concentrations varied widely.[33]

14.Ibrahim A and Maghrabi (2013) were developed **Inductively coupled plasma – atomic emission Spectrometer (ICP-AES)** for the estimation of heavy metals like Cd Pb, K, Ca and Mg in the herbal drugs. The level of the most dangerous heavy metals Cd and Pb in the samples were below the maximum permitted levels reported by world health organization (WHO) standards. K and Ca were present at high levels in samples 2 (Chamomile) and 11 (Becham), respectively. Ca and Mg were most abundant mineral elements in all herbal samples. Moreover, it is observed that the concentrations of most of the tested toxic metals in the investigated herbal plants are found below the permitted levels reported by the international regulatory standards of the medicinal plants.[34]

15.Mousavi Z, Ziarati P,Dehaghi E and Qomi M (2013) were developed **atomic absorption spectrophotometry (AAS)** analytical method for determination of heavy metals such as Pb and Cd in herbal formulation (C-lax, Carvil, Garlet, Galega, Valiflore, Garcine and Ginko)**.** The results indicated that Pb and Cd were present in all investigated herbal drugs. In Conclusion the concentrations of Pb and Cd were higher than the maximum permissible daily levels in the majority of these herbal drugs. [35]

16. Garg M and Singh J(2012) Were developed **Flame atomic absorption spectrometer** and **Atomic absorption spectrometer (AAS)** in which Heavy metals such as Pb, Cd, Ca, Mg, Al, Cu, Zn were eswtimated from 19 popular herbal Churna preparations sold in the Indian market. Heavymetals such as As and Hg were determined by hydride generation technique (cold vapour atomic absorption spectrometery). The results reveal that among the trace (micronutrients) metals Ca and Mg were found in highest amount. Sixteen samples for Hg content and eight for Pb content were exceeding the WHO permissible limits. Arsenic was found below the permissible limit while Cd was above the permissible limit in all the tested samples. In conclusion, the quality of herbal Churna preparations sold in India market is questionable and need to be regulated efficiently before launching in to the market.[36]

17.Kumar G and Gupta Y(2012) were developed **flame atomic absorbance spectroscopy (AAS)** and inductive **coupled plasma–mass spectroscopy (ICP-MS)** for determination of heavy metals such as mercury, arsenic, cadmium and lead in 78 formulations (56 herbal, 19 herbometallic and 3 metallic)Result represented that lead in 19.6% (11/56), cadmium in 21.4% (12/56), mercury and arsenic in 5.3% (3/56) were above the limit. Lead in 52.6% (10/19) of samples, cadmium in 26.3% (5/19) and mercury and arsenic contained in one herbometallic sample was above the limit. Heavy metals in all metal formulations were above the WHO limit. The analytical results of flame AAS and ICP-MS did not differ significantly in the range of measurements in this study, which proves that both methods are satisfactory for estimation of heavy metals in these type of samples.[37]

18.Gajalakshmi S, Iswarya V, Ashwini R (2012) were developed **atomic absorbance spectroscopy (AAS) for** estimation of heavy metals like Ni, Cu, Cr, Zn, Mn, Pb in medicinal plant like Emblicaofficinalis, Phyllanthusemblica, Azadirachtaindica and Balanitesaegyptiaca. The result represent that Emblicaofficinalis, Phyllanthusemblica, Azadirachtaindica, Balanitesaegyptiacahas high level of Cu in it and other metals likeCr, Zn, Mn, Pb are in low amount ,Niand Pb was found completely absent in all the four plants investigated. In conclusion the heavy metal content in the plant was beneficial only to a certain limit.[38]

19.Chris O. Nwoko and Leo Mgbeahuruike (2011) were developed Atomic **Absorption spectrophotometer (AAS)** to determine the heavy metals such as Zn, Cd and Pb in selected ready-to-use herbal remedies (Infecta,Firas , Rinbacin , Koso herbal powder etc). The concentration levels of Pb, Cd were generally high and above the safe limits set by WHO/FAO. Only TunyaB.fil, Virgy worm expeller and Sekin powder had Zn concentrations above international safe limits representing 20% of the tested herbal remedies. The study showed that more than 98% of the Herbal Remedies (HR) analyzed did not comply with international safe limits for herbal products. [39]

20.Yuan X, Ling K and Keung C (2009) were developed **atomic absorption spectrometry (AAS)** analytical method to determine the contents of heavy metals such as As, Pb and Hg contained in Chinese herbal medicine products or Chinese proprietary medicine. Result shows that Limits of detection (LOD) for the three toxic heavy metals were found to be 0.3 ppb for As, 0.1 ppb for Pb and 0.5 ppb for Hg. In Conclusion the developed analytical methods are sensitive enough to detect these heavy metals to meet regulated guidelines. No trace amounts of Hg were found in the test samples due to the fact that the Hg level is less than the LOD. However, variable amounts of As (135.0–5349.3 ppb) and Pb (22.5–968.3 ppb) were found in all products.[40]

21. Gomez M, Cerutti S , Sombra L and Silva M (2007) were developed **Atomic absorption spectrometry (AAS)** analytical method to determine the contents of heavy metals such as Al, Cd, Pb, Fe, Coin Hypericumperforatum leaves and flowers, their teas, tinctures and tablets. The result represent that Cr, and Co were undetectable above their limits of detection in both liquid and solid samples; while aluminum, cadmium, lead, iron and vanadium were present in the majority of samples. In a conclusion the analytical results obtained for all metals indicate that they were present at concentration well below the acceptable daily intake recommended by the World Health Organization.[41]

22. **Caldas E (2003)** were developed **Atomic absorption spectrometry (AAS)** analytical method for estimation of heavy metals such as Cd, Hg and Pb inmedicinal plant like Artichole, Eggplant, Guarana, Horse chestnut, Centella Asiatic, Ginkgo biloba, Ginseng. The result showed that the concentration ofCd exceeding the limit recommended for medicinal plants (WHO). Hgis found much lower than the limit. And Pb is exceeded the maximum recommended limit of WHO. In conclusion The results of this study show the need for a systematic control of toxic heavy metals in plants used as medicines. [42]

23.Bellob M, Ogunwande I, Olawor N (2003) were developed **Atomic absorption spectrometry (AAS)** analytical method to determine the contents of heavy metals such as Mn, Cu, Pb and Zn in herbal plants like Anacardiumoccidentale, Azadirachtaindica, Butyrospermumparadoxum, Mangiferaindica, Morindalucida, Ocimumcanum, Solanumerianthum, Solanumtorvum, Zingiberofficinale and Hyptissuaveolens. The result showed highest mean levels (ppm) of Zn and Cu were found in Hyptissuaveolens while those of Mn and Ca were found in Morindalucida. The result also showed that Ocimumcanum had the highest amounts of Fe. Anacardiumoccidentale had the highest concentration of Na while Azadirachtaindica had the highest mean concentrations of Pb and Mg.[43]

24.CHUANG C, Chen K and HUANG Y (2000) were developed **Atomic absorption spectrometry (AAS)** analytical method to determine the contents of heavy metals such as Pb, Cd, As, Hg, Cu, cobalt, and Mnin Chinese medicines like Panax ginseng, Eugenia caryophyllata, Saposhnikovia divaricate**.** The result represented that cobalt and manganese are generally found in higher concentrations and Cu is usually found in lower concentration and manganese the highest. In conclusion traditional Chinese crude drugs should be warned of the potential danger of heavy-metal poisoning because their concentrations seem to be higher than the maximum values allowed by health agencies in several countries.[44]

5. SUMMARY AND CONCLUSION:

From the review of various literatures, it was concluded that different analytical methods were used as AAS, ICP-MS, GC, EDXRF, FAAS, ICP-OES, and ICP-AES for the analysis of heavy metals such as Pb,Cd, Cur, Fe, Ni, Hg , Cr, Mn, As, and Zn in different

Ayurvedic preparation. . AAS method is simple, convenient and reliable and robust. ICP-MS method was found to be accurate, precise, and rapid. EDXRF method was found to be sensitive, non-destructive and rapid. ICP-AES method was found to be Accurate, precise, and reliable. ICP-OES method was found to be sensitive and rapid. Apart from these HPTLC, TLC, UV method is also used for detection of heavy metals in ayurvedic preparations. These methods were validated by determination of parameters like accuracy, linearity, precision, limit of detection, and limit of quantification as well as robustness and ruggedness. Above method can be used for qualitative and quantitative control for routine analysis of heavy metals in Ayurvedic preparations. These methods can be used for controlling quality control parameters in industries.

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