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DETERMINATION OF SILICON FROM MEDICINAL HERBAL PRODUCT BY ATOMIC ABSORPTION SPECTROPHOTOMETER (AAS), X-RAY POWDER DIFFRACTION (XRPD) AND FOURIER TRANFORM INFRARED SPECTROMETRY (FTIR)

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ABSTRACT

Standardization of Herbal medicines is the challenging work for the pharmaceutical companies. Atomic Absorption Spectrophotometry (AAS), X-ray powder diffraction (XRPD) and Fourier transform infrared spectroscopy (FTIR), are the simple and fast analytical methods. These are powerful tool for the pharmaceutical industry. The technique is used for the phytochemicals analysis of herbal medicines. The present work focused on the qualitative and quantitative analysis of herbal medicine using AAS, XRPD and FTIR methods. Herbal medicine is one of the most popular medicinal system in the world. Electrolytes, Trace elements, Toxic, Essential, Minerals, Alkaloids, Steroids and organic compounds, besides that enzymes, proteins and other Inorganic elements are naturally present in Ayurvedic medicines**.** Popularity of Herbal medicines is growing worldwide because of their minimal side effects. Herbal medicines required standardization, with implementation and constant review of technical standards of production and effective quality control methods. It is necessary to promote this study in the view of the importance of results of both individual and social field. Hence in present study Sarpagandha, Cardiol Vati and Medomine Vati are studied by AAS, XRPD and FTIR for the analysis of Silicon from Herbal medicines.

KEYWORD: Standardization, Herbal medicine, AAS, XRPD, FTIR**,** Silicon.

INTRODUCTION

World Health Organization (WHO) states that around 85-95% of the world population uses traditional Ayurvedic medicines.[1] The poor-quality control of these medicines may cause health hazards like anemia due to destruction of red blood cells. The inorganic elements are needed for metabolism, enzymatic reaction, normal cellular activities and help to maintain the acid-base balance. By using AAS to detect the accurate concentration of inorganic elements. X-Ray Powder Diffraction (XRPD) and FTIR shows the direct information regarding the elements and their physicochemical properties. It is very important technique in the pharmaceutical industry, sensitive method as a phase characterization, polymorphism. Particle size affect the absorption, efficacy of the herbal drugs. Herbal medicinal samples are scanned for the X-Ray powder diffraction by using X-ray diffractometer (Rigaku, Model-Miniflex II) and FTIR. Several modern technologies are used to know the showing 2 θ (°) (Two Theta Degree) value of three strongest peaks of material characterization of Herbal medicines and can be used as fingerprint in quality control of Herbal medicines. Herbal medicine in Ayurveda derived from roots, leaves, fruits, bark, seeds, etc. Various parameters such as dosage, stability, toxicity, chemical factors such as pesticide residues, aflatoxin contents and heavy metals contamination and patient's age (adult or Children) should be taken in to consideration. Airborne toxic effect leading to carcinogenic effect of talcum powder has been accepted by Missouri Jury in a Virginia women case who developed ovarian cancer after decades of using talc -base products. The talc consists of silica derivatives**.** Jury awarded $5**.**4 million in compensatory damages to the lady against Johnson & Johnson company.[2] This shows the severity of Si element. Therefor this study is undertaken to determine Si in Herbal medicines. Most of the people use herbal medicines for less toxicity and minimum side effects. These medicines are now available in different forms like tablets, elixirs, Tonic and powders.[1] Herbal medicines have become more popular as alternative and supplementary remedies in recent years. Contamination or adulteration of herbal medicines with toxic metals, essential elements, trace elements and insect debris[3] are of major concern. The poor-quality control of these medicines causes health hazards like anemia due to destruction of red blood cells. Particle size affect the absorption, efficacy of the herbal drugs, therefor XRPD analysis is essential. World health Organization gives some guidelines[4] for the preparation of herbal medicines and listed some methods for the standardization of herbal medicines[4] and give maximum permissible limit of heavy metals[5] and quality controlled norms. It is important to follow the quality control norms to standardize the herbal medicines. Varies instrumental methods like HPLC -high –performance chromatographic techniques[6],

GC-gas chromatography[6], electrophoresis and TLC -thin layer chromatography[6], XRPD.[7] Are reported for the standardization of herbal medicines maintained the quality and containing well defined constituents are required for reliable beneficial therapeutic effects. Therefor AAS, XRPD and FTIR methods are developed which has high degree of sensitivity and specificity.

MATERIALS AND METHOD FOR AAS

Chemicals

1) Double Deionized water used for all dilution.

2) Hydrogen peroxide (H_2O_2) AR grade (100 Volume, SDFSL.M.W. 34.04).

3) Concentrated Sulphuric Acid (H_2SO_4) (98%).

4) Acetone Extra Pure (Propanone), Assay (GC) min 99.0%.

5) Paraffin liquid heavy (Petrolatum liquid).

Atomic Absorption Spectrophotometer (AAS). Model AA7000F ROM version 1.012. The General analytical conditions are given in table number.

Sampling

In the present study, the marketed herbal tablets of Sarpagandha, Cardiol Vati and Medomine Vati are selected for the analysis. The brand names of products, license number and content as per company's label is included in table 01.

Experimental design

Code numbers namely A, B and D are assigned for above Herbal medicinal samples. By taking the weight of each tablet on digital balance, each tablet sample is gently ground to fine powder using mortar and pestle and packed in butter paper until the analysis. To determine the concentration of Silicon, a wet digestion of the powder sample was done according to the new method developed. Table number 02 shows the weight of Ayurvedic medicinal samples taken for analysis. Each sample is placed separately in 100 mL round bottom flask and 3 mL concentrated sulphuric acid is added. The mixture kept for 30 minutes at room temperature. After 30 minutes 4 mL of 30% hydrogen peroxide is added to the round bottom flask and allowed to cool at room temperature. The sample is then refluxed at 190^0C for 40 minutes. The sample is cooled down to room temperature. 2 mL of 30% hydrogen peroxide is added and the solution heated once again until the digest was clear upon cooling, it is filtered

through Whatman No. 42 filter paper and transferred quantitatively to a 25-mL volumetric flask by adding distilled water.

Sample Preparation and Experimental design for XRPD

By taking the weight of Sarpagandha, Cardiol vati, Medomine Vati, each tablet is gently ground to fine powder using mortar and pestle at room temperature and packed in butter paper until analysis. MiniFlex2 goniometer XRD instrument was set up for analysis.

Sample Preparation and Experimental design for FTIR

A, B and D Code number are assigned for the Herbal samples. Each sample ground to fine powder by using mortar and pestle at room temperature and packed in butter paper. Sample prepared in KBr, small amount of sample individually mixed with KBr.

Table -01. Tablet name with the company name and plants as per label.

Sr. No	Brand and Company Name	Product Name	Plants as per label
1	Baidynath	Sarpagandha	Sarpagandha powder
2	Safe life	Cardiol Vati	Suthi, Arjun ghan, Punarnava, Bringrajn, Abhrak bhasma, shuddha shiljit, Amalki ghan, Guduch ghan, Gokshur ghan, Akik pisti,
3	Safe life	Medomine vati	Pipali, Marich, Amalki ghan, Haritaki ghan, Bibhitaki ghan, Trmad churn, Loha bhasma, Shuddha shilajit, Kitatika, Guduchi, Gugul, Sunthi

Table-02. Weight and dilution of representative Samples for AAS.

Sr. No	Sample Name	Sample Weight(g)	Dilution
1	Sarpagandha (A)	0.336	25 mL in Conc.H_2SO_4
2	Cardiol Vati (B)	0.447	25 mL in Conc.H_2SO_4
3	Medomine Vati (D)	0.448	25 mL in Conc.H_2SO_4

RESULI AND DISCUSSION

Table-03. Elemental concentration in ppm by AAS

Concentration of Silicon in ppm			
Sample	Sarpagandha (A)	Cardiol Vati (B)	Medomine Vati (D)
Silicon	2.00	1.00	3.0

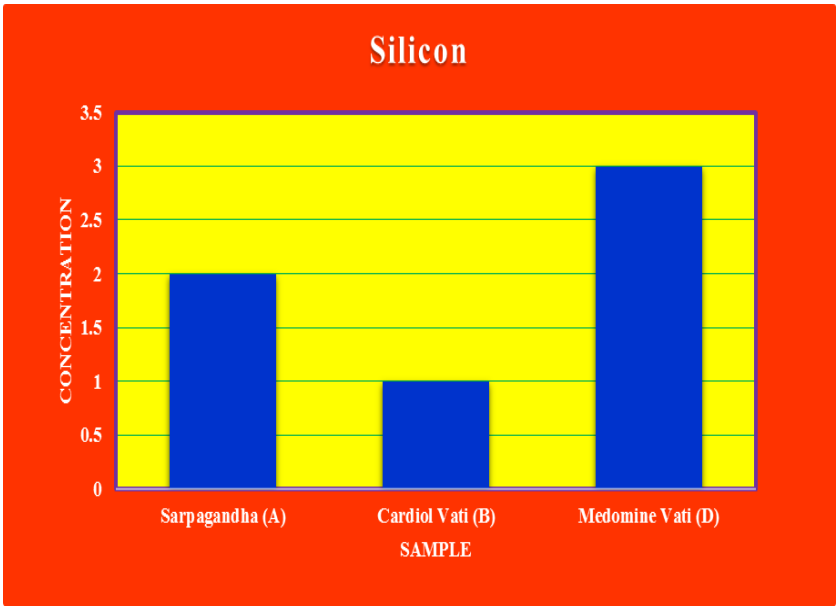

Figure-01 A. Detected Concentration of Silicon present in Herbal Samples

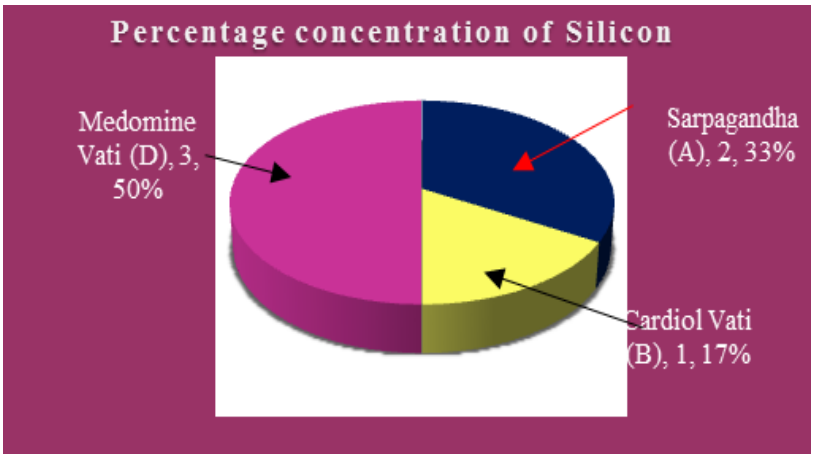

Figure-01B. Percentage Concentration of Silicon present in Herbal Samples

Material for XRPD

Fig-02A. Sarpagandha

Fig-02B. Cardiol Vati

Fig-02C. Medomine Vati

Table-04. Common instrument parameter

Sr. No	Parameter	Values
1	X-ray	Cu / 30 kV / 15 mA
2	Div Slit	1.25 deg
3	Sct Slit	1.25 deg.
4	Rec Slit	0.3mm
5	Scan mode	Continuous
6	Scan speed	5.000 deg./min
7	Sampling width	0.020 deg.
8	Scan axis	2theta/theta
9	Scan range	10.000 -> 80.000 deg.
10	Theta offset	0.000 deg.

XRPD measurements are performed for the sample Sarpagandha (A), Cardiol Vati **(**B**)** and Medomine Vati (D) an analytical instrument with Cu Kα1 radiation with highly sensitive solid state detector. The X-ray tube operated at Cu / 30 kv and 15 mA. XRPD analysis of Sarpagandha tablet, Cardiol Vati and Medomine Vati shows the highest peak at 26**.**48, 26**.**68 and 26**.**64 two theta degree respectively.

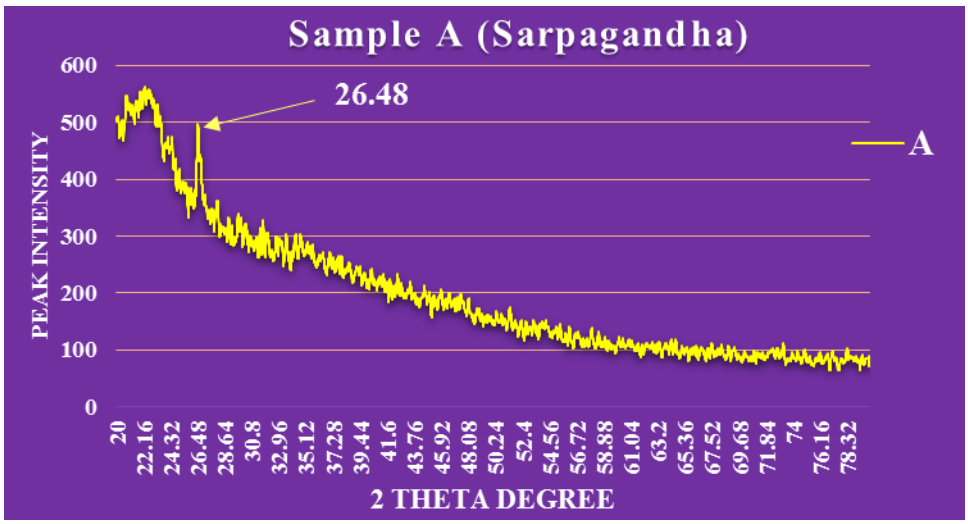

Figure-03. XRPD Graphical representation of Sarpagandha

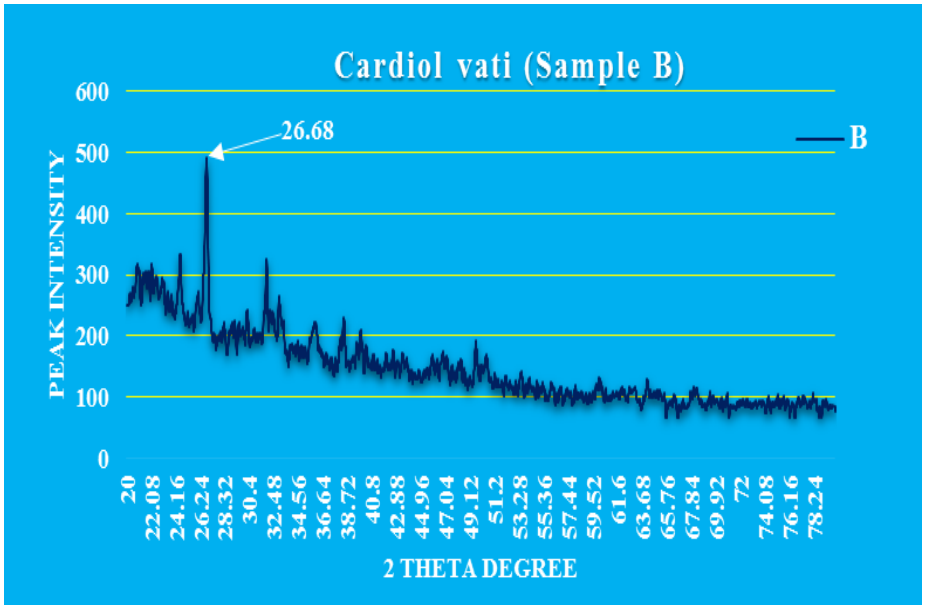

Figure-04. XRPD Graphical representation of Cardiol Vati

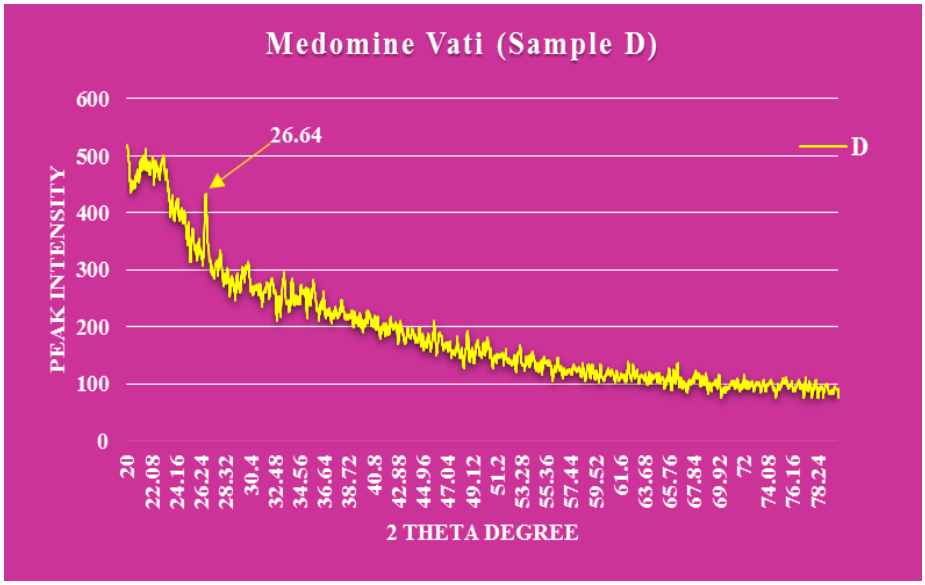

Figure-05. XRPD Graphical representation of Medomine Vati

Table 05- Maximum Intensity and peaks values of Samples at 2 Theta.

Sr. No	Herbal Sample	Two Theta Value	Intensity
1	Sarpagandha	26.48	496.278
2	Cardiol Vati	26.68	460.845
3	Medomine Vati	26.64	426.139

Table 06- Maximum peak value of Standard Silicon Oxide at 2 Theta [8].

Sr. No	Standard	Two Theta Value
1	Silicon Oxide	26.65

Silicon is detected by using modern technique XRPD, X-ray diffractometer indicate the presence of Silicon in all Herbal samples. By using X-ray powder diffractometer to avoid the laborious work and to get instant regarding the presence of inorganic elements.

Material for FTIR

Figure-06A: Sarpagandha

Figure-06B: Cardiol Vati

Figure-06C: Medomine Vati

Instrument "Fourier Transform Infrared Spectra, Perkin Elmer, Model – Spectrum100" is set up for the analysis of Herbal samples. Then sample is placed in the sample holder and positioned in the sample beam of instrument, all spectra were obtained under the identical instrument operating condition. FTIR spectra is recorded in the region of mid-IR region 4000cm^{-1} -400cm^{-1} comparing with standard. The FT-IR spectrum is used as fingerprint to identify the functional group of active compounds based on the peak value on the region of infrared radiation. Rapid quality verification method using FTIR Spectrometer measure the vibration of bonds with chemical functional group and generate a spectrum that can be recorded as biochemical or metabolic "Fingerprint" of the samples.

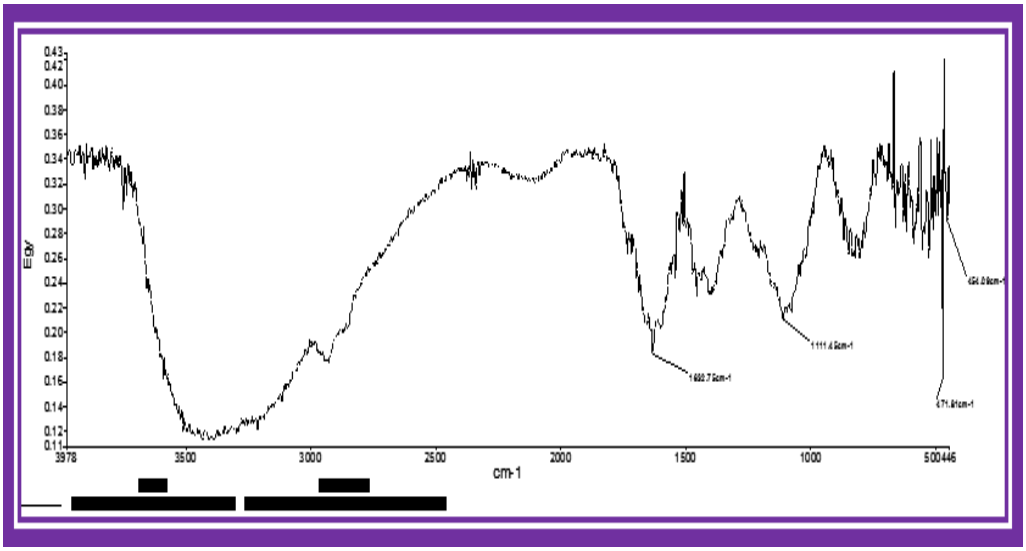

Figure-07. **Sample -A** (**Sarpagandha**)

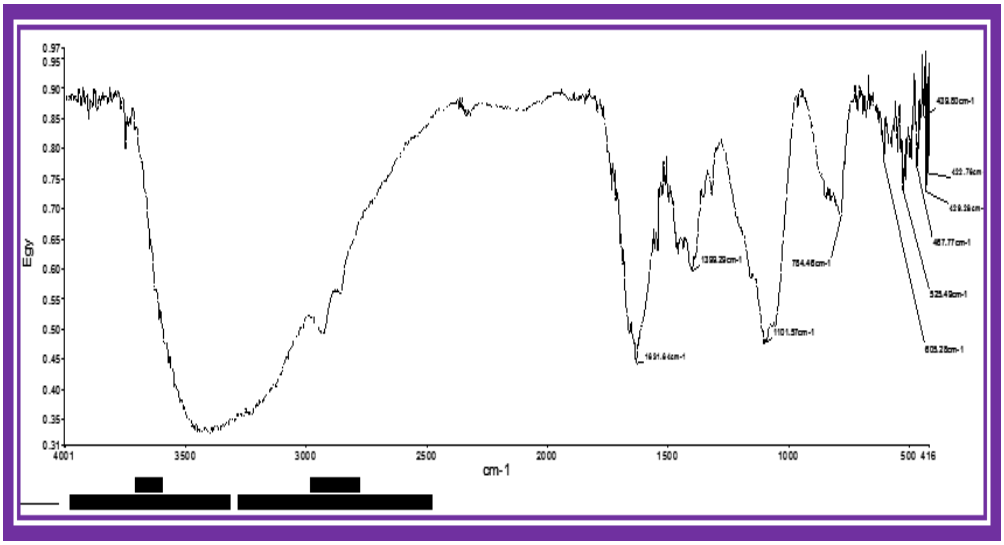

Figure-08. Sample -B (Cardiol Vati)

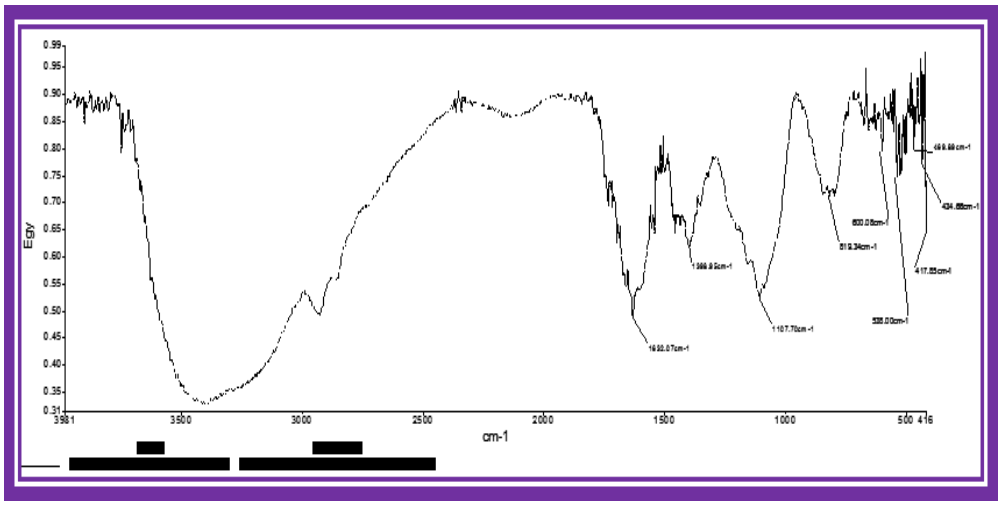

Figure-09. Sample D (Medomine Vati)

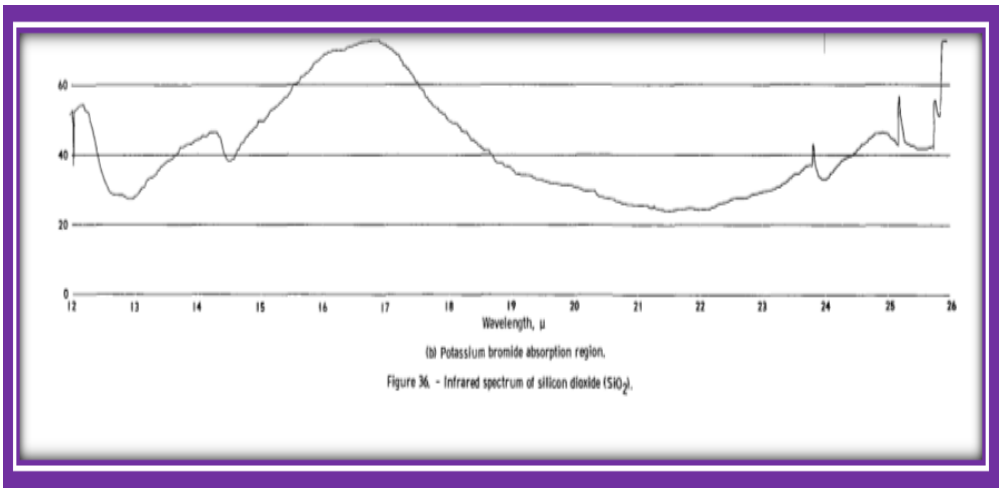

Figure 10- FTIR Spectra for Silicon Oxide as a Standard [9]

Table-07. Frequency for Herbal samples

Sr. No	Frequency for Herbal samples		
	A	B	D
1	1632.75	1631.64	1632.07
2		1399.29	1398.95
3	1111.45	1101.57	1107.70
4			819.34
5		605.28	600.08
6	471.81	467.77	469.89

Molecular vibration due to infrared radiation will be an interesting factor to study in herbal medicines which will have definite physiological effect on human body. The presence of inorganic element in Herbal medicines eventually effects on the stretching or bending on the spectral band and the spectra may be shifted. The spectral data shows that the metal present in most of the Herbal medicines were in the form of metal oxide or in the form of sulphide.

Table-08. Concentration of elements in ppm and LD50, Threshold limit[5,10]

Sr. No	Elements	Samples			LD50	ATL
		A	B	D		
1	**Si**	2.00 ppm	1.00 ppm	3.00 ppm	3.16 mg/kg	10.0 mg/m^3

CONCLUSION

The above study shows that the amount of the element Si present in Herbal sample preparations is very much safe and below toxicity limit for human consumption. Present study shows that effective Ayurvedic ingredients patient consumes are in micro quantities which are in picograms/ fento gram levels per kg body weight. The element Silicon (Si) present in Ayurvedic medicine preparation is below LD50 and air borne threshold limit. FTIR and XRPD are used in the present study can be made mandatory for the quality control of Herbal medicines that can be avoid the laborious and time consuming work.

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