AYUSHDHARA
ISSN: 2393-9583 (P)/ 2393-9591 (O)
An International Journal of Research in AYUSH and Allied Systems

Research Article

ANALYTICAL STANDARDIZATION OF TAMRA YOGA

Rugmini R K[1]*, Sridurga CH[2], Venkata Subbaiah K[3]

*[1]PG Scholar final year, [2]Associate Professor and HOD, Department of Rasa Shastra and Bhaishajya Kalpana, S. V. Ayurvedic College, Tirupati, A.P., India.

[3]Research Scientist, Department of Science and Technology, PURSE centre, S V University, Tirupati, A.P., India.

KEYWORDS: *Tamra Yoga*, Modern Analytical parameters, Safety, Toxicity.

***Address for correspondence**
Dr Rugmini R K
PG Scholar final year,
Department of Rasa Shastra and Bhaishajya Kalpana,
S. V. Ayurvedic College,
Tirupati.
Email: rkrugmini@gmail.com
Cell: 9966619437

ABSTRACT

Rasa Shastra is a specialized branch of Ayurveda which mainly deals with the pharmaceutics of unique and potent preparations. *Tamra Yoga* is an important *Rasa Oushadi* mentioned in Rasa Tantra Sara Va Siddha Prayoga Sangraha which contains *Tamra Bhasma, Yashtimadhu, Chincha Kshara, Trikatu, Sauvarchala lavana* and *Hingu*. *Shodhana, Bhavana, Marana, Amrutikarana, Chincha Kshara nirmana* and *Churna nirmana* are the main pharmaceutical procedures employed in the preparation of *Tamra Yoga*. To assess the toxicity, safety and to understand the structural and chemical composition, it was tested through various modern analytical parameters like X-ray diffraction (XRD), Scanning electron microscopy (SEM), Energy dispersive X-ray spectroscopy (EDS), Particle size analysis (PSA), Zeta Potential (ZP), UV-Spectroscopy, Fourier transform Infra-Red spectroscopy (FTIR) and Inductively Coupled Plasma – Optical Emission Spectrometry (ICP-OES). XRD of *Tamra Yoga* shows major peaks of KCl (Potassium Chloride), CuS (Copper Sulphide) and minor peaks of HgS (Cinnabar), NaCl (Sodium Chloride), CaS (Calcium Sulphide) and ZnP_4 (Zinc Phosphide), $K_2Fe_2O_4$ (Potassium Iron Oxide). SEM micrographs showed an agglomeration of crystalline irregular sharped particles; EDS analysis confirmed the significant presence of elements viz. O-27.91%, S-21.83%, Cu-26.87% and Hg-14.29%, K- 3.46%; Particle size was found to be 337.9nm and its Zeta Potential is -12.1mV. UV- Spectrum of *Tamra Yoga* showed maximum absorption at 307 nm; FT-IR analysis showed 11 peaks between the wavelengths 3356.21 - 418.34 cm^{-1} and ICP–OES analysis revealed Potassium as main constituent in 14376.50 ppm.

INTRODUCTION

Analytical study plays an important role in the standardization of the drugs. Ayurveda, the ancient system of medicine is gaining recognition throughout the world and many herbal, metal and mineral drugs are now clinically tested and accepted. However, one of the impediments in the acceptance of the ancient systems of medical preparation is the lack of standard quality control profiles. The quality of the drugs, that is, the profile of the constituents in the final product has implication in efficacy and safety.

Tamra Yoga is one of the important Herbo-metallic formulations mentioned in Rasa Tantra Sara va Siddha Prayoga Sangraha - Prathama Khanda – Bhasma Prakarana[1]. It contains one part of *Tamra Bhasma* and four parts of *Chincha Kshara* (Alkali of *Tamarindus indica* fruit rind), *Hingu* (*Ferula foetida*), *Yashtimadhu* (*Glycyrrhiza glabra*), *Trikatu* and *Sauvarchala Lavana* (Unaqua Sodium Chloride) each. All the *Dravyas* possess various therapeutic properties indicated in the management of several diseases. Classical texts have enumerated certain tests which indicate the proper transformation of basic metal into bio-absorbable *Bhasma* form. In spite of these facts, *Bhasmas* of metals are always under debate, not

only in sense of its therapeutic excellence but also for unnecessary hue and cry about their toxicity and safety. Therefore modern analytical techniques are expected to help in circumventing this problem. Hence highly sensitive modern parameters like X-Ray Diffraction, Scanning Electron microscopy, Energy Dispersive X-Ray Spectroscopy, Particle size analysis, Zeta Potential, UV-Spectroscopy, Fourier transform Infra-Red spectroscopy and Inductively Coupled Plasma - Optical Emission Spectrometry were employed for gaining information about identity, form, particle size, surface morphology, structure and contents of the formulation.

MATERIALS AND METHODS

Parada, Gandhaka and *Tamra* were obtained from the local market of Vijayawada. *Chincha Phala Twak* was obtained from Adilabad district of Telangana State. Hingu, Souvarchala Lavana, *Yashtimadhu* and *Trikatu* were obtained from TTD's Sri Srinivasa Ayurveda Pharmacy, Tirupati. Entire preparation of *Tamra Yoga* was carried out in Department of Rasa Shastra and Bhaishajya Kalpana, TTD's S.V.Ayurvedic College and Sri Srinivasa Ayurveda Pharmacy, TTD, Tirupati. Requirement for XRD: Model- Powder X-Ray Diffractometer D8 advance, Manufacturer- Bruker Germany. SEM and EDS: Model- EVO MA 15, Manufacturer- Carl Zeiss - Germany; PSA and ZP: Model- Horiba scientific Partical Size and Zeta Potential Analyzer, Manufacturer- Horiba instruments, Irvine, CA 92618 USA; UV-Spectroscopy: Model- Nano drop 8000 Spectro-photometer, Manufacturer- Thermo Scientific, India; ICP-OES: Model- Agilent 725, Manufacturer- Agilent technologies, USA.

Pharmaceutical process

The pharmaceutical procedures adopted in this study are *Shodhana, Bhavana, Marana, Amrutikarana, Kshara nirmana, Hingu bharjana*, *Churna nirmana* and preparation of capsules of *Tamra* Yoga. *Shodhana* of *Parada* was done by *Mardana* with *Kshara traya* (*Sarja Kshara, Yava Kshara, Tankana*) for three days[2]. *Shodhana* of *Gandhaka* was performed by *Puta* method using cow's milk[3]. Equal quantities of *Shodhitha Parada* and *Gandhaka* were taken and made into *Kajjali*[4]. *Tamra Patras* were subjected to *Samanya shodhana* by *Nirvapa* in *Taila, Takra, Gomutra, Aranala* and *Kulattha Kwatha* for seven times[5]; *Visesha shodhana* was done by *Dola yantra swedana* in Gomutra for three hours[6]. Equal quantities of *Kajjali* and *Shodhitha Tamra Patras* were triturated in a *Khalwa yantra* using *Nimbu Swarasa. Chakrikas* of uniform size were prepared and placed in a *Sharava* and subjected to *Sharava samputikarana*. This was subjected to *Laghu puta* and the entire procedure was performed for 18 times[7]. Then the *Tamra Bhasma* having all the *Bhasma lakshnas* have been attained. Then they obtained *Tamra Bhasma* was triturated with *Kumari Swarasa* and subjected to *Amrutikarana* procedure by *Laghu puta* for 7 times[8]. *Chincha phalatwak* was taken and converted to ash by heating in a mesh placed over the hearth. To the ash obtained four parts of water was added and kept overnight. Then the supernatant water was collected and heated in a moderate flame. *Chincha Kshara* was obtained[9]. Raw drugs of *Yashtimadhu, Sauvarchala lavana, Trikatu* and *Hingu* were made into fine powder. Then one part of *Tamra Bhasma* and 4 parts each of *Chincha Kshara, Yashtimadhu churna, Sauvarchala lavana churna, Trikatu churna* and *Hingu churna* were mixed together to prepare *Tamra Yoga* and the homogenous mixture were filled in capsules of 655mg.

Analysis of *Mandura Bhasma* using ancient parameters (*Bhasma Pariksha*)

The final *Bhasma* was analyzed for quality control as described in the ancient texts and the following observations were made:

- Rekhapurnatva[10]: After proper trituration, small amount of *Bhasma* was taken between thumb and index finger. It filled into the fine lines of fingers. *Rekhapurnatwa* was obtained after 14th *Puta*.
- *Varitaratwa*[11]: After proper trituration, small amount of *Bhasma* was sprinkled on the surface of water. *Bhasma* being light floated on the surface of water. This was obtained after 18th *Puta*.
- *Nischandratwa*: Small quantity of *Bhasma* was observed under bright sunlight for presence of any free shiny metal particle. There was no shining particle observed in the *Bhasma* after 3rd *Puta*.
- *Niswadu Pareeksha:* When a small amount of the *Bhasma* was kept on tongue, there was not any feeling of taste / untoward sensation.
- *Dantagrenakachkachabhavati*: When a small amount of the *Bhasma* was placed between the teeth, no sandy feeling was appreciated.
- *Anjanasadrishyasukshmatva*: The *Bhasma* prepared was fine like collyrium.
- *Avami*: Ingestion of small amount of the *Bhasma* did not produce any nausea / vomiting.
- *Amla Pareeksha: Tamra Bhasma* was taken in little quantity and sprinkled over the curd taken

in a watch glass and kept undisturbed for 24 hours. No bluish discolouration was seen after 24 hours.

- *Nimbu Swarasa Pareeksha*: Very little quantity of *Tamra Bhasma* was added to the fresh *Nimbu Swarasa* taken in a test tube and kept aside for 24 hours. On the next day there was no colour change in the lemon juice.
- Discolouration was not found in *Dadhi Pareeksha* and *Amla Pareeksha* after 16th *Puta*.

Analysis of *Tamra Yoga* using modern parameters

X-Ray Diffraction (XRD)

Tamra Yoga was subjected to XRD at Department of Physics, Yogi Vemana University, Kadapa, Andhra Pradesh.

Procedure: Sample was powdered in agate mortar to very fine powder and it was mounted in sample tray of machine. X-Ray beam bearing a wavelength of 1.5418740 A° from copper source is passed on the sample. Detector was set to identify diffracted beams between 10 -70 degrees of 2 range. Obtained soft files of XRD consisting values of 2θ and intensity are plotted on a graph (2θ on X-Axis and Intensity of Y-Axis) using "Origin Pro 8.5 SR2" Data Analysis Software. Various compounds consisting similar diffraction pattern were identified by matching their peaks with corresponding JCPDS Crystallographic cards. For even better accuracy and precision, XRD soft files were also analyzed for corresponding phase/entry matching with Crystallographic Open Data base (COD - 20120320) – USA, after plotting values in PANalytical X'pert high score plus software 3.0.0.123, UK.

Scanning Electron Microscopy and Energy dispersive X-Ray spectroscopy

The practical was performed at Department of Physics S.V University, Tirupati.

Procedure of SEM: Specimen of the sample to be analyzed was directly kept on the specimen holder for visualization. As the sample employed has nonconductive nature, the sample surface was coated by carbon using arc melting technique. Then the dried powder was observed under the microscope at 1,000X to 10,000KX and the micrographs were taken with the inbuilt camera.

Procedure of EDS: Electron beam excitation is used in scanning electron microscopes (SEM). A detector is used to convert X-ray energy into voltage signals; this information is sent to a pulse processor, which measures the signals and passes them on to an analyser for data display and analysis. The detector used in EDS is often the Lithium drifted Silicon detector which is operated at liquid nitrogen temperatures. Sample of *Tamra Yoga* was placed on the specimen holder and subjected to Energy-Dispersive X-ray spectroscopy (EDS). When the sample was bombarded by the SEM's electron beam, electrons are ejected from the atoms comprising the sample's surface. The resulting electron vacancies are filled by electrons from a higher state, and an X-ray is emitted to balance the energy difference between the two electron's states. The X-ray spectrum thus acquired gives the information on the elemental composition of the material under examination.

Particle Size Analysis and Zeta Potential

The practical was conducted at Department of science and Technology, PURSE, S.V.University, Tirupati.

Procedure of PSA: The sample was mixed in water and sonicated for 10 minutes. Then it was poured into the sample chamber, where it passes through the laser beam as homogeneous stream of particles. The scattering of light occurs over a wide range of angles upon interacting with the particles in the suspension which are moving by Brownian motion. Based on this scattering pattern of sample, particle size distributions are calculated comparing with appropriate optical model.

Procedure of ZP: A 1% concentration of *Tamra Yoga* was prepared in distilled water. The particles were well dispersed before analysis and the sample was taken in a 1ml syringe and injected slowly into the capillary cell (cuvette) through the sample port. Care was taken to see that air bubbles are not formed during this process. As the sample comes out from the second port of the capillary cell, the injection process is stopped. This indicates complete filling of the sample into the capillary cell. The sample ports are then covered with lids and the capillary cell was then placed into the sample holder of the zeta sizer instrument for analysis.

UV- Spectroscopy

Practical was performed at Department of science and Technology, PURSE, S.V.University, Tirupati.

Procedure: 5gm of *Tamra Yoga* was macerated with 100 ml of solvent in a closed flask for twenty-four hours, shaking frequently during six hours and allowed to stand for eighteen hours. It was filtered, taking for UV spectroscopic study. The Spectra was taken at 200-800 nm from the peak obtained, the λmax value was calculated.

Fourier Transform Infrared Spectroscopy (FT-IR)

This practical was conducted at Padmavathi Mahila University, Tirupati.

Procedure: Sample was placed in the Potassium bromide plate of FTIR spectrometer and the interference pattern was detected by the infrared detector as variations in the infrared energy level, and the obtained spectral information was calculated.

Inductively Coupled Plasma – optical Emission Spectrometry

This practical was performed at Centre for material for electronics technology (C-MET), department of Electronics and Information technology, Hyderabad.

Procedure: 0.2 g of *Tamra Yoga* was taken in Teflon tubes and added to 6.0 ml of Nitric acid and 2.0 ml of Hydrogen peroxide and allowed for 10 minutes in outside for reaction. Then samples were dissolved using Microwave Digestion System (Anton PaarMultiwave 3000). Then the *Tamra Yoga* solutions were made to 25.0 ml and filtered. These solutions were used for Elemental analysis using ICP-OES instrument.

RESULTS

X-Ray Diffraction Studies (XRD)

Table No. 1: Showing the details of matching peaks of XRD data for *Tamra Yoga*

S.No	Element/Molecule	JCPDS Ref.No	2θ	Intensity	FWHM	h	K	L
1.	KCl (Potassium Chloride)	00-004-0587	28.30	100	0.24	2	0	0
2.	HgS (Cinnabar)	01-075-1589	26.42	85	0.384	1	0	1
3.	HgS (Cinnabar)	00-042-1408	31.19 43.59 51.74	93 21 13	0.24 0.48 0.48	1 1 2	0 1 0	2 0 1
4.	CuS(Copper Sulphide)	00-024-0060	31.76 52.67 58.60	100 23 13	0.072 0.288 0.24	1 - -	0 - -	3 - -
5.	NaCl (Sodium Chloride)	00-005-0628	66.22	6	0.384	4	0	0
6.	CaS (Calcium Sulphide)	00-001-0980	45.30	100	0.168	2	2	0
7.	ZnP_4 (Zinc Phosphide)	00-022-1015	47.94	30	0.384	-	-	-
8.	$K_2Fe_2O_4$ (Potassium Iron Oxide)	00-002-0802	31.58	100	0.096	2	2	0

Table No. 2 Showing Crystal details of JCPDS entries:

Name	KCl
Space group	Fm3m
Crystal System	Cubic
Cell Parameters	a = 6.2931 A˚ b= 6.2931 A˚ c = 6.2931 A˚
Z	4.00

Name	HgS (01-075-1589)
Space group	P3121
Crystal System	Hexagonal
Cell Parameters	a = 4.1600 A˚ b= 4.1600 A˚ c = 9.5400 A˚
Z	3.00

Name	HgS (00-042-1408)
Space group	P3121
Crystal System	Hexagonal
Cell Parameters	a = 4.1495 A˚ b= 4.1495 A˚ c = 9.4970 A˚
Z	3.00

Name	CuS
Space group	P63/mmc
Crystal System	Hexagonal
Cell Parameters	a = 3.7960 A˚ b= 3.7960 A˚ c = 16.3600 A˚
Z	6.00

Name	NaCl
Space group	Fm3m
Crystal System	Cubic
Cell Parameters	a = 5.6402 A˚ b= 5.6402 A˚ c = 5.6402 A˚
Z	4.00

Name	CaS
Space group	Fm3m
Crystal System	Cubic
Cell Parameters	a = 5.6860 A˚ b= 5.6860 A˚ c = 5.6860 A˚
Z	4.00

Name	$K_2Fe_2O_4$
Space group	-
Crystal System	Cubic
Cell Parameters	a = 7.9900 A˚ b= 7.9900A˚ c = 7.9900 A˚
Z	4.00

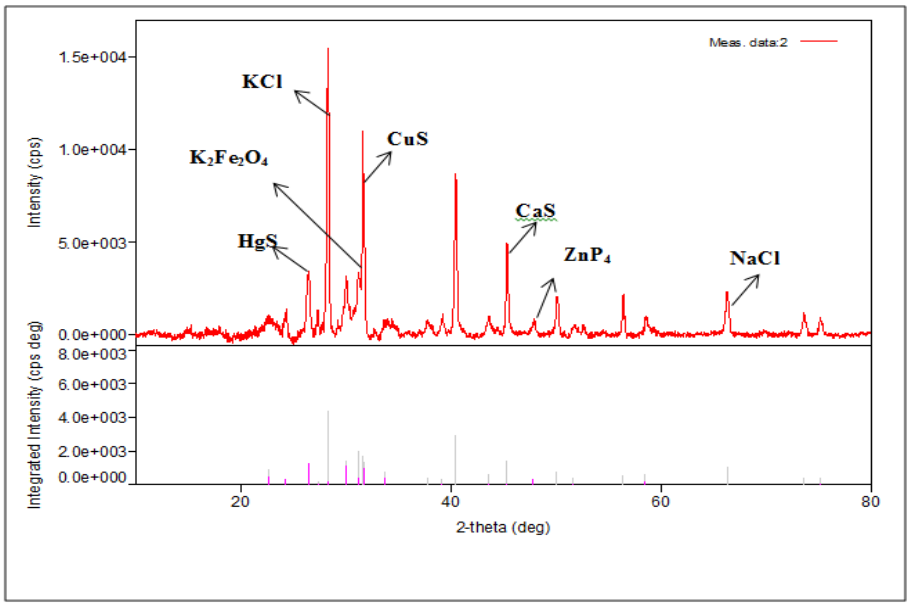

Figure No.1: Showing XRD graph of *Tamra Yoga*

XRD of *Tamra Yoga* shows major peaks of KCl (Potassium Chloride) compound with cubic structure and CuS (Copper Sulphide) compound with hexagonal structure. Minor peaks showed the presence of HgS (Cinnabar) compound with hexagonal structure, NaCl (Sodium Chloride) and CaS (Calcium Sulphide) with cubic structure, ZnP_4 (Zinc Phosphide), $K_2Fe_2O_4$ (Potassium Iron Oxide) with cubic structure. KCl peak was detected at diffraction angle of 28.30, CuS peaks at 31.76, 52.67, 58.60, HgS peaks at 26.42, 31.19,43.59, 51.74, NaCl peak at 66.22, CaS peak at 45.30, ZnP_4 peak at 47.94 and $K_2Fe_2O_4$ peak at 31.58diffraction angle.

Scanning Electron Microscopy:

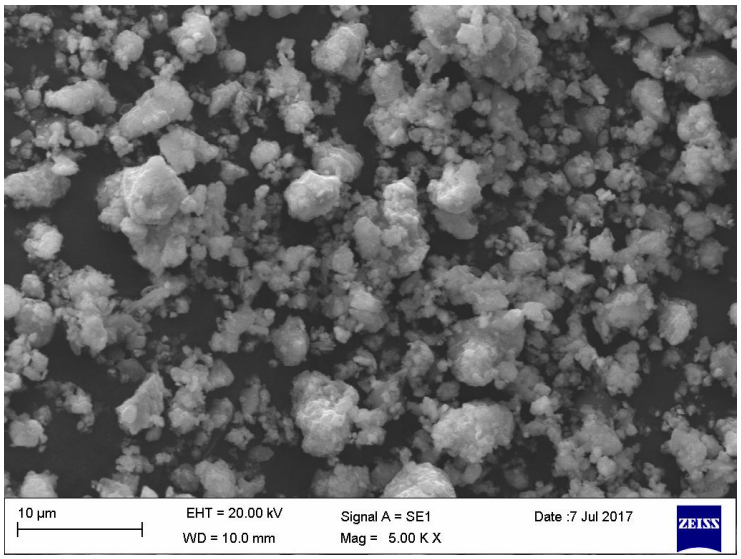

Figure No.2: Showing SEM result of *Tamra Yoga* (Mag. 5KX)

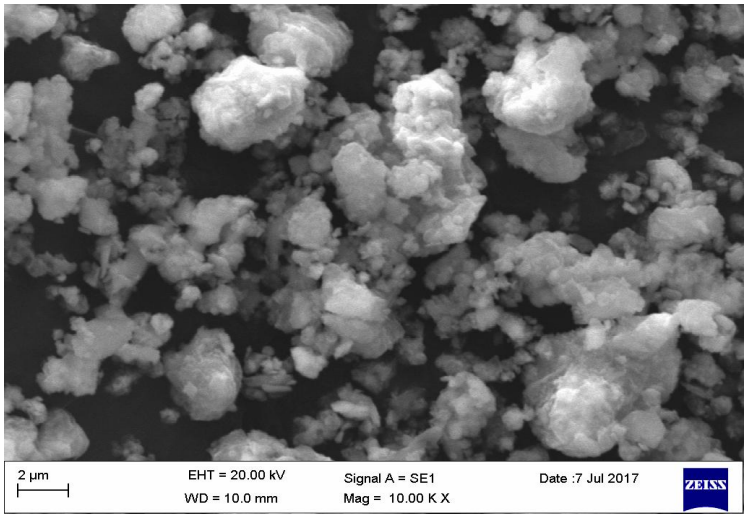

Figure No. 3: Showing SEM result of *Tamra Yoga* (Mag. 10KX)

SEM micrographs of *Tamra Yoga* showed an agglomeration of crystalline irregular sharped particles in a disordered proportion at 5KX and 10KX magnifications.

Energy Dispersive X-Ray Spectroscopy

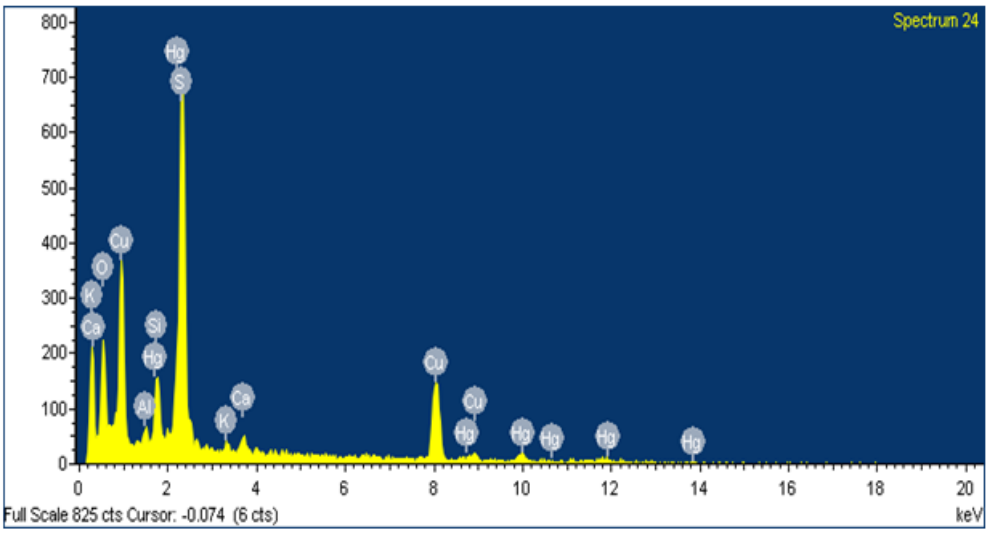

Figure No.4: Showing EDS graph of *Tamra Yoga*

Table No. 3: Showing the quantity of all the elements in *Tamra Yoga*

Element	Weight%
O K	27.91
Mg K	0.90
Al K	0.71
Si K	3.17
S K	21.83
K K	3.46
Fe K	0.87
Cu K	26.87
Hg M	14.29

EDS analysis of *Tamra Yoga* confirmed the presence of elements viz. O- 27.91%, Mg- 0.90%, Al- 0.71%, Si- 3.17%, S-21.83%, K-3.46%, Fe-0.87%, Cu-26.87%, Hg-14.29%.

Particle Size Analysis

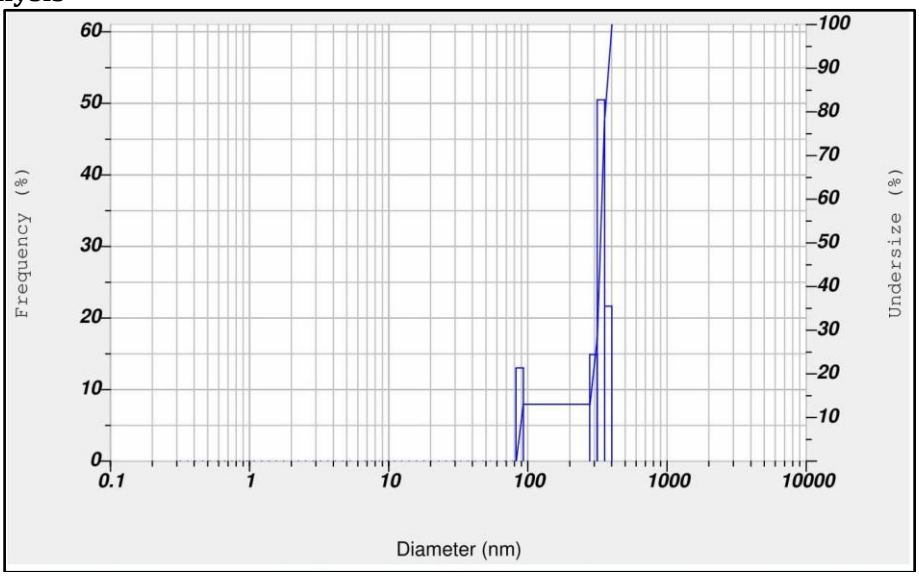

Figure No.5: Showing the result of Particle size analysis of *Tamra Yoga*

The mean particle size of *Tamra Yoga* is **337.9 nm.**

Zeta Potential:

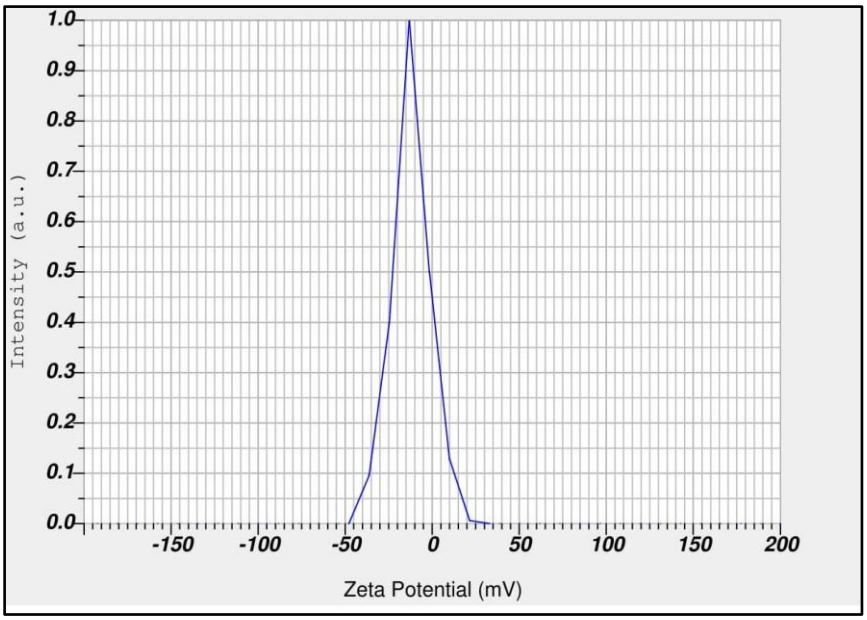

Figure No. 6: Showing Zeta potential distribution of *Tamra yoga*

Tamra Yoga sample showed a Zeta potential value of **-12.1mV,** which indicates low colloidal stability.

UV- Spectroscopy

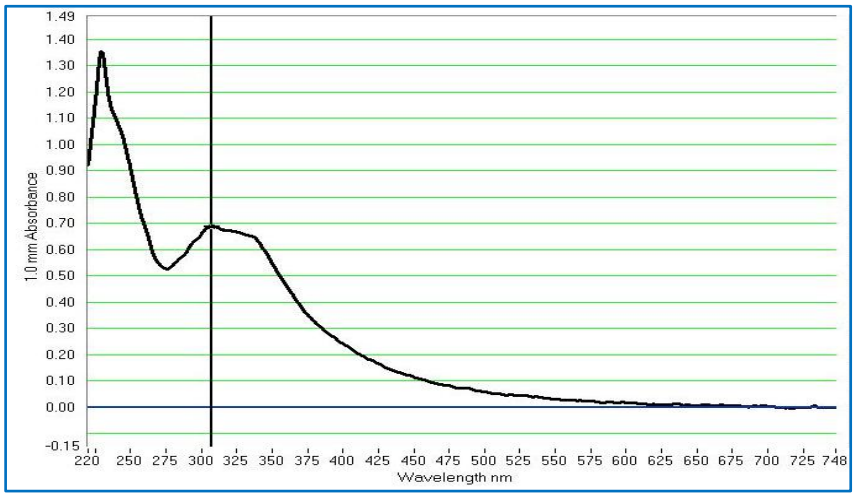

Figure No. 7: Showing UV-Spectrum of *Tamra Yoga*

UV - Spectrum of *Tamra Yoga* showed maximum absorption at **307 nm.**

Fourier Transform Infrared Spectroscopy (FT-IR)

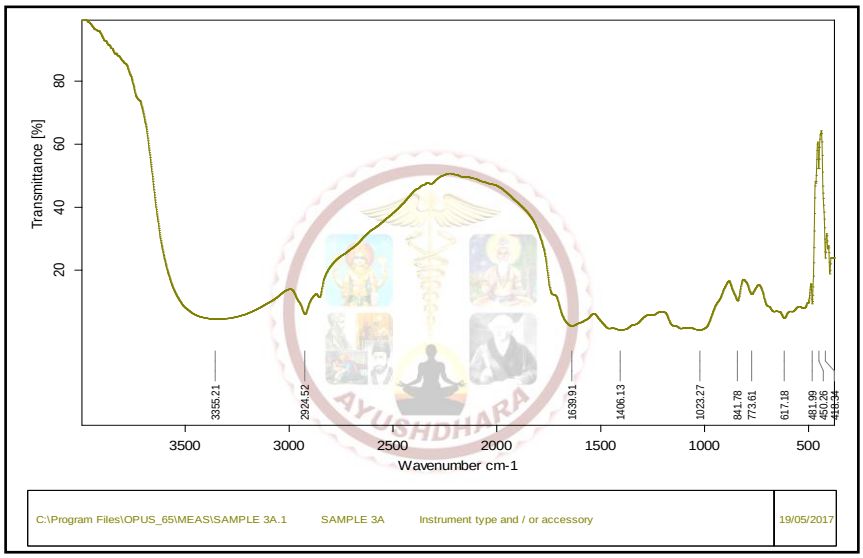

Figure No.8: Showing various peaks obtained in FTIR analysis of *Tamra Yoga*

Table No.4: Showing details of Peaks obtained in FTIR analysis of *Tamra Yoga*

Sample Name	No. of Peaks	Wavelength
Tamra Yoga	11	3356.21, 2924.52, 1639.91, 1406.13, 1023.27, 841.78, 773.61, 617.18, 481.99, 450.26, 418.34.

Table No.5: Various peaks obtained in FTIR analysis of *Tamra Yoga* and their correlation with compounds

S.No.	Peak	Actual peak	Bond	Type of bond	Appearance
1.	3200-3400 cm^{-1}	3356.21	O – H	Alcohols, Phenols	Broad
2.	2850-3000 cm^{-1}	2924.52	C – H	Alkane	Strong
3.	1620-1680 cm^{-1}	1639.91	C = C	Alkene	Variable
4.	1000-1400 cm^{-1}	1406.13 1023.27	C – F	Alkyl Halide	Strong
5.	675-1000 cm^{-1}	841.78 617.18 773.61	=C – H	Alkene	Strong

Inductively Coupled Plasma – optical Emission Spectrometry

Table No.6: Showing the result of ICP-OES analysis of *Tamra Yoga*

S.No.	Name of the elements analyzed	Tests results in ppm
1.	Silver	3.49
2.	Arsenic	ND
3.	Boron	21.24
4.	Calcium	1233.16
5.	Cadmium	ND
6.	Chromium	12.01
7.	Copper	6374.84
8.	Iron	401.90
9.	Mercury	1650.22
10.	Potassium	14376.50
11.	Magnesium	976.78
12.	Manganese	64.62
13.	Sodium	449.36
14.	Phosphorus	432.49
15.	Lead	93.14
16.	Sulphur	6264.10
17.	Selenium	ND
18.	Tin	14.18
19.	Zinc	239.86

DISCUSSION

Analytical study is a process which helps in identification of quantitative and qualitative data of a substance, the components of a solution or mixture, or the determination of the structures of chemical compounds. It is an essential part of any thesis work. It gives us the knowledge about identity, size, structure of chemical constituents and physical properties. It hints us about toxic properties of drugs, if any.

X-Ray diffraction has been in use in two main areas, for the finger print characterization of crystalline materials and the determination of their structure. Each crystalline solid has its unique characteristic X-Ray powder pattern, which may be used as a "fingerprint" for its identification. Once the material has been identified, X- Ray crystallography may be used to determine its structure, i.e. how the atoms pack together in the crystalline state and what the inter-atomic distance and angle etc. X-Ray diffraction is one of the most important characterization tools used in solid state chemistry and material science. Size and the shape of the unit cell for any compound can be detected most easily using the diffraction of X-rays. Major peaks of KCl and CuS, minor peaks of HgS, NaCl, CaS, ZnP_4, and $K_2Fe_2O_4$ were found in the XRD of Tamra Yoga.The major peaks formed were sharp indicated crystalline nature of KCl and CuS. The presence of minor peaks may be due to the formation of organo metallic complex. These peaks formed were not as sharp as those of CuS and KCl. Presence of KCl in the major peak of *Tamra yoga* may be due to the presence of higher amounts of potassium in *Chincha Kshara* reacting with NaCl present in the *Sauvarchala Lavana* (black salt). HgS occurs in two forms cinnabar and metacinnabar. Formation of cinnabar requires a temperature of more than 270°C, while metacinnabar forms at temperature ranging from 20°C - 90°C. Hence, we can justify the formation of Cinnabar from the heat produced due to *Laghu puta* (514^0C). The shape of crystals was found to be hexagonal. Copper and sulphur reacts at higher temperatures in the absence of oxygen resulting in the formation of Copper Sulphide (CuS).

Scanning electron microscopy (SEM) is an analytical technique to know the surface morphology of the drug. It uses electron beam rather than light to form a Figure. It is capable of producing high resolution figures of a sample surface, which means that closely spaced features can be examined at a high magnification. Due to the manner in which the Figure is created, SEM Figures have a characteristic three dimensional appearance and are useful for determining the surface structure of the sample i.e. topography. It can magnify objects to extreme levels where even structure of nano

particles could be clearly visible. Agglomerations of crystalline irregular sharp particles were seen in micrographs of *Tamra yoga*. This agglomeration in the surface morphology of *Tamra Yoga* may be due to the presence of natural tannins, resins and salts in the component drugs with binding nature, which may be sticking to the surface of *Tamra Bhasma*. The bigger particles look like agglomeration of small particles. Presence of huge amounts of various multi elemental organic molecular peaks in XRD evidences this phenomenon.

Energy-Dispersive X-ray spectroscopy (EDX) is an analytical technique used for elemental analysis or chemical characterization of a sample. It relies on the investigation of an interaction of some source of X-ray excitation and a sample. EDS of *Tamra Yoga* revealed the presence of elements like Oxygen, Copper, Sulphur, Mercury and Potassium. The presence of other elements like Silica, Iron and Aluminium may be due to addition of herbal ingredients.

The size of the particles in the drug plays major role in its therapeutic action and efficacy. Particle size and surface area of solid drug are inversely related to each other. The mean particle size of *Tamra Yoga* is 337.9 nm. The nano size of drug is indicative of its quick absorption and faster dispersion into body resulting into better therapeutic efficacy. Zeta potential is a measure of the magnitude of the electrostatic or charge repulsion or attraction between particles, and is one of the fundamental parameters known to affect stability. The Zeta Potential (mean) value of *Tamra Yoga* found to be -12.1 mV which indicates its low colloidal stability. High zeta potential indicates easy dispersion, whereas less zeta potential indicates strong aggregation of particles in suspension. Particle aggregation refers to formation of assemblages in a suspension. During this process, particles dispersed in liquid phase stick to each other. This occurs by addition of salts or other chemicals referred to coagulant or flocculent. Low colloidal stability of *Tamra Yoga* may be due to the presence of *Sauvarchala lavana* and *Chincha Kshara*.

UV-Spectroscopy refers to absorption spectroscopy or reflectance spectroscopy in the ultraviolet-visible spectral region. Different molecules absorb radiation of different wave-lengths. An absorption spectrum will show a number of absorption bands corresponding to structural groups with the molecule. Electromag-netic spectrum of U.V region is from 190 to 400 nm whereas for visible region it is 400-800 nm. UV-Spectrum of *Tamra Yoga* showed maximum absorption at 307 nm which shows its absorbency in UV region.

FTIR was performed to detect the presence of functional groups or organic legends in *Tamra Yoga*. Infrared spectroscopy deals with the infrared region of the electromagnetic spectrum that is light with a longer wavelength and lower frequency than visible light. When infrared light or radiation hits a molecule, the bonds in the molecule absorb the energy of the infrared and respond by vibrating. *Tamra Yoga* showed 11 peaks between the wavelengths 3356.21 - 418.34 cm^{-1}. Strong intensity of C-H stretching and C-F stretching vibrations resulted in one peak at 2924.52 cm^{-1} and two peaks at 1406.13 and 1023.27 cm^{-1} which were assigned to Alkane and Alkyl halide respectively. A peak at 3356.21 cm^{-1} raised due to O-H stretching vibrations represents alcohols and phenols. Three peaks were obtained between the wavelengths 675-1000 cm^{-1} due to =C - H stretching vibrations indicate the presence of Alkenes. Variable peak obtained near 1639.91cm^{-1} represents C = C stretching vibrations. This indicates that there are no complex structures in *Tamra Yoga*.

ICP-OES is one of the most powerful and popular analytical tool for the determination of trace elements in a sample. It is very useful for standardization as well as to develop analytical profile. ICP–OES analysis of *Tamra Yoga* showed Potassium as main constituent in 14376.50 ppm. This may be due to the presence of potassium in *Chincha Kshara, Maricha, Pippali* and *Yashtimadhu*. 6374.84 ppm of Copper, 6264.10 ppm of Sulphur and 1650.22 ppm of Mercury indicates the presence of *Tamra Bhasma*. Calcium is present in 1233.16 ppm. This may be due to the presence of calcium in Trikatu and Yashtimadhu[12]. Sodium in 449.36 ppm confirms the presence of *Sauvarchala lavana*. Zinc, Magnesium and Phosphorous were present in 239.86, 976.78 and 432.49 ppm respectively. These minerals are available in *Nimbu Swarasa*[13], *Kumari*[14], *Yashtimadhu*[15], *Trikatu*[16] and *Hingu*[17]. Toxic elements like Arsenic and Cadmium were not detected.

CONCLUSION

From the present study, it can be confirmed that *Tamra Yoga* is a herbo metallic compound with the presence of KCl and CuS as the major phase. Nano particle of the yoga indicates its quick absorption, dissolution to the target site and high therapeutic efficacy; presence of other micro elements may be due to the other herbal ingredients used in the formulation. Free metal or toxic elements were not identified; this indicates

the proper transformation of *Tamra* into bioavailable form (*Bhasma*). Complete transformation of a base metal into *Bhasma* is the prime requisite to avoid any adverse effects and toxicity. Ancient analytical parameters like *Bhasma pariksha* help in only understanding the qualitative nature, whereas modern analytical parameters reveal the structural characterisation as well as the chemical composition. Hence both the parameters must be used for the justification of the formulation, for a safe therapeutic approach.

REFERENCES

1. Rasa Tantra Sara Va Siddha Prayoga Sangraha, Rajasthan: Krishna Gopal Ayurveda Bhavan, 2013, p-49.
2. Pandit Kasinath Shastry, Sri Sadanandasharma Virachitha Rasa Tarangini, Taranga 5 Verse no. 34-35, Varanasi: Motilal Banarasidas, 2014; p-81.
3. Dr. Parimi Suresh, Rasendra Sara Sangraha of Krishna Gopalabhatt, Chapter 1, Verse no: 120-121, Varanasi: Choukhambha Sanskrit Samsthan, 2012, p-44.
4. Pandit Kasinath Shastry, Sri Sadanandasharma Virachitha Rasa Tarangini, Taranga 2 Verse no. 27, Varanasi: Motilal Banarasidas, 2014; p-16.
5. Dr. Indradev Tripati, Sri Vagbhatavirachitha Rasa Ratna Samucchaya, Chapter 5, Verse no.13, Varanasi: Choukhambha Sanskrit Samsthan, 2013; p-53.
6. Dr. Indradev Tripati, Sri Vagbhatavirachitha Rasa Ratna Samucchaya, Chapter 5, Verse no. 52, Varanasi: Choukhambha Sanskrit Samsthan, 2013; p-58.
7. Dr. Indradev Tripati, Sri Vagbhatavirachitha Rasa Ratna Samucchaya, Chapter 5, Verse no. 53, Varanasi: Choukhambha Sanskrit Samsthan, 2013; p-58.
8. Pandit Kasinath Shastry, Sri Sadanandasharma Virachitha Rasa Tarangini, Taranga 17 Verse no. 43-44, Varanasi: Motilal Banarasidas, 2014; p-419.
9. Prof.K.R.Srikantha Murthy, Sarangadhara Samhitha, Madhyama Khanda Chapter 11, Ver.102-104, Varanasi: Choukhambha Orientalia, 2010; 156.
10. Dr. Indradev Tripati, Sri Vagbhatavirachitha Rasa Ratna Samucchaya, Chapter 8, Verse no.28, Varanasi: Choukhambha Sanskrit Samsthan, 2013; p-90.
11. Dr. Indradev Tripati, Sri Vagbhatavirachitha Rasa Ratna Samucchaya, Chapter 8, Verse no.27, Varanasi: Choukhambha Sanskrit Samsthan, 2013; p-90.
12. Sampat Ghosh and Manu Singh, Minerals and Heavy metals contents of Glycyrrhiza glabra L. and Andrographis Paniculata (BURM.F) from Meerut, India by, International Journal of Pharma and Biosciences, 2015, Oct;6 (4): (P) 40-45.
13. Abdelhafeez M.A. Mohammed, Amna M. Ibrahim, Ayat A. Omran, Moawia E. Mohamed, Saumaya E. Elsheikh, Minerals content, essential composition and physicochemical properties of Citrus jambhiri lush. (Rough Lemon) from the Sudan, International Letters of Chemistry, Physics and Astronomy;9(1); 2013; 25-30.
14. Amar Surjushe, Resham Vasani, DG Saple; Aloe vera: A short review; Indian journal of Dermatology 2008; 53(4): 163-166.
15. Sampat Ghosh and Manu Singh, Minerals and Heavy metals contents of Glycyrrhiza glabra L. and Andrographis Paniculata (BURM.F) from Meerut, India by, International Journal of Pharma and Biosciences, 2015, Oct;6 (4): (P) 40-45.
16. Olubunmi B. Ajayi, Seun F. Akomolafe and Funmilayo T. Akinyeni, Food value of two varieties of Ginger commonly consumed in Nigeria, International Scholary Research notices nutrition, Volume 2013, Article ID 359727, 5 pages.
17. Shah N.C and Amir Zare, Asafoetida (Heeng): The well-known medicinal condiment of India and Iran by, The Scitech journal; April 4(1); 2014; 30-36.

Cite this article as:
Rugmini R K, Sridurga CH, Venkata Subbaiah K. Analytical Standardization of Tamra Yoga. AYUSHDHARA, 2017;4(5):1297-1307.

Source of support: Nil, Conflict of interest: None Declared