Research Article

Analytical Evaluation of An Ayurvedic Formulation - Abhraka Bhasma

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ABSTRACT

Abhraka Bhasma is called as a wonder drug due to its curative property in various ailments. Hence there is an urgent requirement to prove its non-toxicity and therapeutic value and also to conform genuinity and safety before administration to human beings. In this study, Abhraka Bhasma is authenticated as biotite mica by observing quality assurance methods and testing it through standard ayurvedic and analytical techniques like EDXRF, FEG-SEM and EDS. For this purpose commercially available Abhraka Bhasma was procured and subjected to conventional method of analysis wherein the sample satisfied all the criterias given in the Ayurvedic Pharmacopoeia. Bhasma proved to be brick red in colour, lusterless, fine, had floating character, was excellently prepared (Unama test), tasteless, without alloy, smooth and tangible. The modern analytical method EDXRF revealed the presence of Fe (22%) as a major element and Ca, K and Si in low concentrations, their concentration being 11%, 8% and 13% respectively. Mg (4%), Al (2%) and Ti (1%) were present as minor elements while Sodium, Chlorine, and Phosphorous were present in traces (<1%). FEG-SEM studies showed that the grains in Abhraka Bhasma were heterogenous and in aggregates of particle size between 19 nm and 88 nm. The grains were found to be irregular in shape ranging from spherical to oblong. EDS analysis show that major elements present in the sample) were O (41%), Si (16%), K (13%) and Fe (13%) and the minor elements were Al (6%), Mg (5%), Ca (4%) and Cl (1%). Sodium, Phosphorous and Titanium were found in traces (<1%). The analytical techniques used confirmed the absence of organic compounds and mercury in the sample characterized. This physico-chemical characterization study will help to take stock of Bhasma preparations as novel nano-technological applications giving a new thrust to Ayurvedic pharmaceutics.

Keywords: Biotite, Pharmacopoeia, Unama test, tangible, FEG-SEM, mercury.

INTRODUCTION

Mineral drugs (*Rasa Yoga*), developed by the branch of Ayurveda known as 'Rasashastra', are the products of Ayurvedic metallurgy. These formulations take on different valence states and have different crystal structure and physical properties. It is obvious from the literature that the use of metallo-mineral drugs, which are subsequently formed after progressive blooming of *Sodhana*, *Bhavana* and *Marana* processes, are considered pharmaceutically most suitable forms.

Abhraka Bhasma is the Bhasma of the mineral, mica (Biotite) [$K(Mg,Fe)_3(AlSiO_{10})(OH)_2$]. Its synthesis involves repeated calcinations which transforms the metallic state into corresponding oxide form. Calcination is repeated 10 cycles (dasa puta), 100 cycles (satha puta), 1000 cycles (sahastra puta), etc.. Going by its brick red colour, the end product is iron oxide, probably Fe_2O_3. Biotite shows up with FeO and Fe_2O_3.[1] FeO oxidizes to Fe_3O_4 and possibly to Fe_2O_3 also. The only purpose large number of calcinations serve is for achieving fine scale particle sizes.

Abhraka Bhasma is widely used in cases of pernicious and sickle cell anemia, Bells Palsy, hepatic dysfunction, leukemia, sex debility, azoospermia, cystic fibrosis, post encephalic dysfunction and cervical dysplasia to name few. Its primary use is as aphrodisiac, astringent and restorative (powerful cell regenerator). It is also known for its penetrative and spreading property in the whole body and various micro tissues. It is an alternative, aphrodisiac, anti-pyretic, carminative, hematinic and proven rejuvenator.

Dosage of the drug is as follows: [2]

- For 5 to 10 years – 120 mg
- For 11 to 25 years – 240 mg
- Above 25 years – 360 mg

In Ayurvedic system, there is dearth of information of solid scientific evidence regarding safety, efficacy, quality of practices and precise molecular mechanism of Abhraka Bhasma. Elemental composition is very often overlooked in biochemical assays. It is essentially required to discuss its non-toxicity and therapeutic value and to conform genuinity and safety before administration to human beings. During the Bhasma making, the end products is made in batch sizes of unprecedented kind. Strict adherence to the processes as delineated in the classical texts ensured that the finished product was as expounded. Different methods of Bhasma preparation as given in the classical texts fail to reproduce authentic 'Rasaoushadhies'. The tests to check the formation of the Bhasma described in the literature are raw and designed with the limitations of developed science in that age. Further, some of these tests are highly individualistic.

With the advent of modern technology it thus becomes imperative to put the synthetic procedures used in making Abhraka Bhasma under scrutiny using modern instruments. So as also to resolve the respective indications and strengthen the regime to monitor the manufacturing and administration of this preparation, analytical study becomes imperative. Adulteration/ substitution as well as presence of free particles will have an effect on the quality and safety of the drug. It becomes obligatory to adopt modern analytical methodology to determine the important chemical constituents present in the drug qualitatively and quantitatively. This will make understanding and interpretation of pharmacological action of any drug easier and better. Hence, this study was conducted and aims at analytical evaluation of Abhraka Bhasma on structural and elemental basis and to address the role of it. This study will provide preliminary data about this Ayurvedic preparation; it being important, not only for standardization and consumer protection, but also in advancing drug research.

For material characterization of the Abhraka Bhasma sample, the fingerprints of the Bhasma sample was generated by using Rasa shastra quality control tests like *Nishchandratva, Rekhapurnatvam, Varitaratvam*, *Amla pariksha, Nisvadutam* etc. A systematic characterization of this traditional drug was done by using various state-of-the-art modern techniques viz., Energy dispersive X-ray fluorescence (**EDXRF**) spectrometer, Field Emission Gun - Scanning Electron Microscopy (**FEG-SEM**) and Energy Dispersive Spectroscopy (**EDS**) as analytical analyzer.

MATERIALS AND METHODS

Sahasraputi Abhraka Bhasma (subjected to 1000 putas) was procured from a renowned organization, Shree Dhootapapeshwar Ltd, Khetwadi, Mumbai, India.

Classical specification techniques for standardization

While taking into account the quality control measures for the standardization of Rasaoushadhies, following preliminary analysis of Abhraka Bhasma were performed. These ancient methods as prescribed by AYUSH were included in the study to assure the reliability in terms of safety and efficacy of the products. Ayurvedic texts have described methods for quality control of finished products through different parameters like *Nischasndratva*, *Varitara*, *Nirutha*, *Apunarbhava* etc.. [3]

The procured Abhraka Bhasma was analyzed as described in Ayurvedic texts as follows and found suitable:

1. *Nishchandratva:* The *chandrika (luster*) present in the metal is due to its physical property, which reflects the light when fall on it. The absence of chandrika in the Bhasma indicates that every particle of the mineral has been incinerated and converted in to Bhasma form. Abhraka Bhasma was taken in a petri dish and visually examined for any luster in bright sunlight through magnifying glass. No luster was observed in the Bhasma i.e. it's incineration was complete.
2. *Rekhapurnatvam*: It is an organoleptic method conducted by the fingertips to study *sukshmata* (fineness) of any Bhasma. Bhasma particles should be of minimum size for penetration of the Bhasma particles up to minute capillaries of the body without any obstruction, best absorption and assimilation in the body. The rate of absorption of drug is directly proportional to the particle size of drug.[4] Finer the particle size, quicker is the absorption. A pinch of Abhraka Bhasma was taken in between the thumb and index finger and rubbed. It was observed that the Bhasma fill the lines and crevices of the finger easily, and was not washed out from the cleavage of the lines i.e., the Bhasma was *rekhapurnatva* (fine). It is a common parameter to be applied for any product/formulation, which contains drugs of mineral/metal origin. Abhrak Bhasma passed this test.
3. *Varitaratvam*: Varitara test, applied to study lightness and fineness of Bhasma, is floating character of Bhasma on stagnant water surface. This test is based on the law of surface tension. A small amount of the procured Abhraka Bhasma was sprinkled over the still water in a beaker. It was found that the Bhasma particles floated over the surface of the water i.e., the Bhasma was *varitaratva* (floats on water). This indicates that particle size of the Bhasma does not break the surface tension of the water and also uniformity of the particles. [5]
4. Unama test: It is a further assessment of Varitara test. A grain of rice was kept carefully on the layer of floated Bhasma. The grain remained as it is on the layer indicative of its excellent preparation.
5. *Nisvadutam*: The prepared Bhasma was found to be tasteless when a small amount was kept on the tongue i.e., the Bhasma was *nisvad* (tasteless).
6. *Amla pariksha*: A pinch of prepared Bhasma was mixed with a little amount of *dahi* (curd) in a clean and dry petridish and observed for any color change. No color change of *dahi* was observed. The same procedure was followed with lemon juice taken in a clean and dry test tube, and the same result was observed. This test confirms that the sample was not an intermediate but that the Bhasma had been obtained.
7. *Avami*: Ingestion of 5-10 mg of the Bhasma did not produce any nausea/vomiting.
8. *Nirdhoom*: Small quantity of the Bhasma was heated and no smoke was produced i.e., it was *nirdhoom.*
9. *Niruttha*: Rajat test involved heating of a very thin silver sheet (600 nm thickness) along with a small quantity of Bhasma to red hot for about 5 minutes. After self cooling the sheet to room temperature, no traces of this sample stuck permanently to the silver sheet. Further, weight of silver was taken. There was no increase in weight of silver leaf which indicated that Bhasma was properly prepared and there was no alloy formation. This confirmed that the metal had

totally transformed into its oxide form constituting the Bhasma and that the Bhasma was irreversible.

10. *Anjana Sannibha : Anjana* (coryllium) is smooth in character and it does not create any irritation whenever applied. Abhraka Bhasma was found to be smooth without creating any irritation to mucous membrane i.e., it was properly incinerated.
11. *Slakshnatvam*: It is a sensation produced by Bhasma by simple touch with finger tips. Properly incinerated Bhasma attain this quality. *Slakshna Bhasma* can be absorbed and assimilated in the body without producing any irritation to mucous membrane of gastrointestinal tract. Abhraka Bhasma was found to be tangible.
12. *Dantagreakachikachitatva:* In this test when the Bhasma was put in to the mouth and pressed in between teeth there was not a typical '*kach-kach*' sound. It means the particle was that much fine that there were no hard or sand like materials.
13. V*ishishtavarnotpatti:* After Marana process there should be an emblematic color development in the end product. That color should match to the mentioned color in the classical reference of that procedure. The sample drug was of brick red in color in accordance with the text.[6]

The conventional tests like *Varitara*, *Rekhapurna*, *Nirittha* etc. performed to check the quality of Bhasma are not quite reliable. Characterization of Abhraka Bhasma using modern analytical tools becomes inevitable. Hence, the Bhasma was further analyzed using modern technical instruments.

Modern analytical techniques for standardization

After the Bhasma complied with Rasa shastra quality control tests, the Bhasma was analyzed using analytical instruments – EDXRF, FEG-SEM and EDS. An attempt has been made to establish the sensitivity of the basic requirement of a scientific study on this material with respect to the manufacturers of Bhasmas, Ayurvedic practitioners as well as end-users.

EDXRF: The EDXRF analysis was conducted for a quantitative scan of the test drug sample using EDXRF spectrometer [Make: HORIBA-Japan; Model: MESA-500W]. The specifications of the instrument were as follows:

Live Time: 100 sec	Quant. Corr.: Standardless
X-ray tube vo.: 15/50 kV	Current: 360/46 microA
Path: Vacuum	CELL: Nonexistence

FEG-SEM: The surface characteristics and morphology of the Bhasma sample was determined by FEG-SEM (JEOL, JSM 7600F, Japan) with PC-SEM software. Since, our main aim of this work was to achieve a consistent submicrometer-scale characterization of the mica nanoparticles, we have applied advanced, high-resolution, Field-Emission-Gun, Scanning Electron Microscopy (FEG-SEM).

In order to get a higher quality secondary electron image for FEG-SEM examination, a representative portion of the sample was sprinkled onto a double-sided carbon tape. For coating it with an ultrathin electrically conducting material, gold/palladium alloy was deposited on the sample by SPUTTER COATING on Hummer V sputter coater. The micrographs were obtained with a secondary electron detector at an accelerating voltage of 4 V and probe current of 10 A for 3 mins. This fixation was found necessary to eliminate sample drift and sample damage under the electronbeam at very high magnifications, which were necessary to detect mica nanoparticles.

For mounting the sample, the coated sample was placed inside the microscope's vacuum column through an airlock chamber. An electron beam was thermionically emitted from an electron gun fitted with a tungsten filament cathode at the top. Thermally assisted Schottky type Field Emission Guns (FEG) using electron emitters of zirconium oxide were used.

This beam travelled downward through a series of magnetic lenses designed to focus the electrons to a very fine spot. Near the bottom, a set of scanning coils made the focused beam to move back and forth across the mounted sample, in a raster fashion. As the electron beam hit each spot on the sample, secondary electrons were emitted from its surface. A detector counted these electrons and sent the signals to an amplifier. Accelerating voltage was 10kV. The final image was built up from the number of electrons emitted from each spot on the sample.

EDS: A quantitative compositional analysis was carried out on Inca analyzer EDS spectrometer at SAIF/CRNTS, IIT, Mumbai. Low-voltage EDS approach was used by analysing the Fe-L and Pd-L low-energy spectral lines.

RESULTS

Energy-dispersive X-ray Spectroscopy (EDXRF)

The typical concentration of elements present in the sample as determined by EDXRF analysis for this sample is tabulated in Table 1.

This additional phase analysis revealed the presence of Fe (22%) as a major element and Ca, K and Si in low concentrations, their concentration being 11%, 8% and 13% respectively. Mg (4%), Al (2%) and Ti (1%) were present as minor elements while Sodium, Chlorine, Phosphorous were present in traces (<1%), presence of which could be attributed to the various herbal products used during the heat treatments of the Maran procedure. The product does not contain any free metal even at trace levels. This was found to be acceptable with respect to Ayurvedic pharmacopeia. The investigation also revealed that elements were present in oxide forms, only 0.10% of KCl was detected in the sample. No carbon present in it indicates the absence of any natural organic matter. The weight (%) of several elements taken together indicates the presence of the predominant oxide group with mainly Si and Fe as ionic species.

Field Emission Gun-Scanning Electron Microscopy with (FEG-SEM)

FEG-Scanning Electron Microscopic studies showed that the grains in Abhraka Bhasma were non-uniformly arranged (heterogenous) in aggregates of particle size between 29 nm and 88 nm. The grains were found to be irregular in shape ranging from spherical to oblong. Thus it may be concluded that mica gets converted into a mixture of simple compounds having very small particle size after the particular process of Marana. This is the first report of fingerprinting of Abhraka Bhasma. It is worth noting that FEG-SEM micrographs displayed no impurities.

Electron Dispersive Spectroscopy (EDS)

Chemical composition of Abhraka Bhasma with omission of peaks in 5 and 8 keV range:

Spectrum processing:

Peaks possibly omitted: 5.900, 8.025, 8.607

KeV Processing option: All elements analyzed (Normalised)

Number of iterations = 4

Table 1: The typical concentration of elements present in the sample as determined by EDXRF analysis

Element	Line	Mass [%]	Standard Deviation 2 sigma [%]	Intensity [cps/microA^0]	Formula	Mass [%]
12 Mg Magnesium	K	3.522	0.339	0.042	MgO	5.839
13 Al Aluminium	K	2.36	0.185	0.102	Al2O3	4.459
14 Si Silicon	K	12.543	0.205	1.058	SiO2	26.832
19 K Potassium	K	7.844	0.203	1.854	K2O	9.449
20 Ca Calcium	K	10.698	0.245	2.657	Cao	14.968
22 Ti Titanium	K	1.452	0.143	1.582	TiO2	2.422
11 Na Sodium	K	0.114	0.047	0.235	Na2O3	0.166
26 Fe Iron	K	21.534	0.282	60.973	Fe2O3	30.788
17 Cl Chlorine	K	0.08	0.03	0.24	KCl	0.1
15 P Phosphorous	K	0.218	0.036	0.794	P2O5	0.273
8 O Oxygen		35.392	0.41			

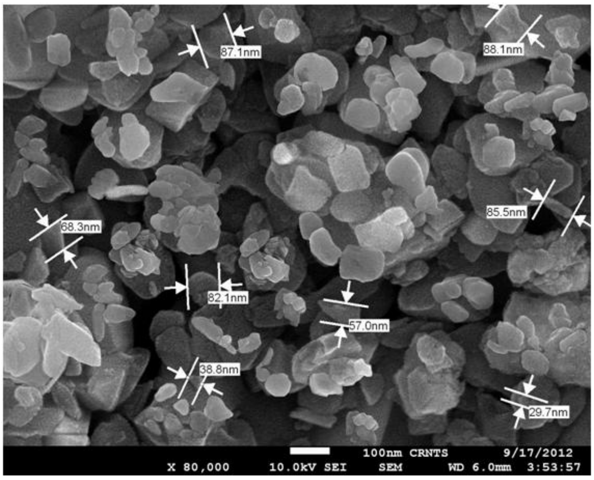

Figure 1: Field Emission Gun- Scanning Electron Microscopic features of Abhraka Bhasma

EDS analysis was used to analyze elemental composition of the samples. Table 2 depicts the chemical composition of Abhraka Bhasma with omission of 5.900, 8.025, 8.607 keV peaks. The results show that major elements present in the sample (after rounding off) were O (41%), Si (16%), K (13%) and Fe (13%) and the minor elements present were Al (6%), Mg (5%), Ca (4%) and Cl (1%). Sodium, Phosphorous and Titanium were found in traces (<1%). This is well supported by spectra shown in Fig.2.

Table 2: Chemical composition of Abhraka Bhasma with omission of 5.900, 8.025, 8.607 keV peaks

Element	Weight %	Atomic %
O K	40.53	59.25
Na K	0.51	0.52
Mg K	4.95	4.76
Al K	6.19	5.36
Si K	15.94	13.27
P K	0.45	0.34
Cl K	0. 31	0.86
K K	13.1	7.84
Ca K	4.73	2.18
Ti K	0.78	0.38
Fe K	12.52	5.24
Totals	100	

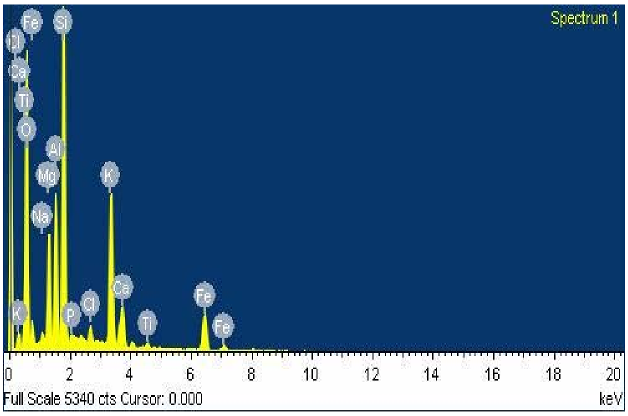

Figure 2: Spectra of Abhraka Bhasma with omission of 5.900, 8.025, 8.607 keV peaks

Chemical composition of Abhraka Bhasma with omission of peaks in 8 keV range

Spectrum processing:

Peaks possibly omitted: 8.034, 8.625 keV

Processing option: All elements analyzed (Normalised)

Number of iterations = 4

Table 3: Chemical composition of Abhraka Bhasma with omission of 8.034, 8.625 keV peaks

Element	Weight %	Atomic %
O K	42.36	60.7
Na K	0.64	0.64
Mg K	6.56	6.19
Al K	3.93	3.34
Si K	14.98	12.23
P K	1.59	1.17
S K	0.81	0.58
Cl K	0.81	1.17
K K	8.02	4.7
Ca K	9.16	4.67
Ti K	0.73	0.35
Fe K	10.42	4.28
Totals	100	

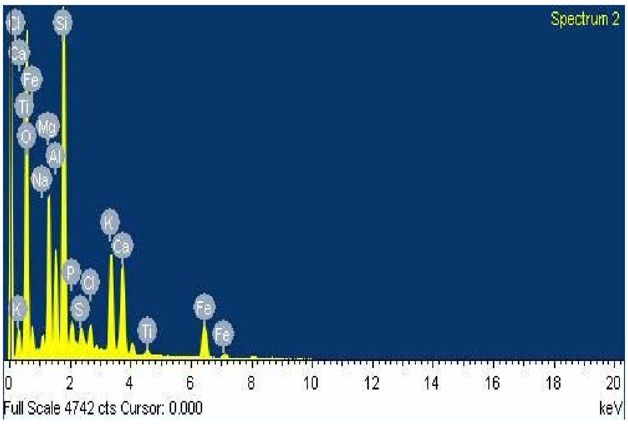

Figure 3: Spectra of Abhraka Bhasma with omission of 8.034 and 8.625 keV peaks

When the peaks omitted were in the range of 8 keV alone, the results showed that major elements present in the sample (after rounding off) were O (42%), Si (15%), K (8%) and Fe (10%) and the minor elements present were Al (4%), Mg (7%) and Ca (9%). Chloride, Sodium, Phosphorous and Titanium were found in traces (<1%) (Table 3). This is well supported by spectra shown in Fig.3.

DISCUSSION

Standardization is a measurement for ensuring the quality and is used to describe all measures, which are taken during the manufacturing process and quality control leading to a reproducible quality. The Bhasmas have all along been a mystery, as a substance and as a medicine, the reason being that the end product may sometimes be unstable. The lack of understanding of traditional methods resulted in a difficulty to reproduce authentic preparations. In view of such ambiguity and the risk due to their inconsiderate use, there is an urgent need to bring about a standardization of the preparation process and the end product. Dixit (1987) first gave the scientific basis for standardization of Bhasma.[7] Standardization of Bhasma is utmost necessary to confirm its identity and to make sure the safety, effectiveness and acceptability of the product.

But the most important challenges faced by these formulations are the lack of complete standardization by physico-chemical, microbiological and analytical evaluation.[8] Very few reports are available where attempts have been made to understand the physico-chemical properties of Bhasma.

Abhraka Bhasma is a wonder drug of Ayurvedic Pharmacopoeia well known on virtue of its therapeutic properties. Hence its analytical study is necessary to find out adulterisation/substitution as well as presence of free particles that will have an effect on the quality and safety of the drug. The commercial sample of Abhraka Bhasma satisfied the required criteria of traditional standardization techniques. However, the classical qualitative tests do not provide any information about the composition and the structure of the final drug. [9] There is an urgent need of scientific methods for assessing the quantitative and structural informations. An attempt has been made to summarize three important analytical techniques, EDXRF, FEG-SEM and EDS available for standardization of Abhraka Bhasma.

The modern standardization techniques EDXRF confirmed the presence of metals such as Fe, Al, Mg, Fe, K, Si and Ca along with few trace elements (presence of which in the drug can be considered as negligible). The weight (%) of several elements taken together indicates the presence of the predominant oxide group i.e., Abhraka Bhasma particles are in oxygen efficient state. The results indicate that oxides of Silicon, Potassium, Calcium and Iron were predominantly present in comparison to other elements constituting Abhraka Bhasma. Fe_2O3 had the highest weight percent in the sample (30.8%), followed by SiO_2 (26.9%). The quantitative estimation of the major and minor elements present in Abhraka Bhasma were within the standard limits given for Abhraka Bhasma by Pharmacopoeial standards for Ayurvedic Formulations.[10]

The SEM pictures, with x 80,000 resolution (10 kV) of Abhraka Bhasma particles were taken which suggested its size and morphology. The images revealed that the surface of the Abhraka Bhasma particles, are in nanoparticle range with atleast one dimension < 100nm in diameter. It confers to the definition of nanoparticles according to which "nanoparticles typically have at least one dimension less than 100nm in size".[11] FEG-SEM suggested that the observed particles were aggregates and were found to be irregular, heterogenous and of sizes varying from 29 nm to 88 nm. From all these observations it is evident that sample Abhraka Bhasma consists of particles of various sizes and shapes. Thus, FEG-SEM can assist in proper structural characterization of of Ayurvedic medicines.

EDS has been used to detect the elements as weight % and atomic mass % present in the sample. Table 2 and Table 3 shows the weight % and atomic % of different constituents of Abhraka Bhasma. Figure 2 and Figure 3 shows spectra of Abhraka Bhasma using EDS with different spectrum processing techniques. The weight % of Sodium, Phosphorous, Chlorine and Titanium (when the peaks omitted were in the range of 5 and 8 keV) were found to be 0.51 %, 0.45 %, 0. 31% and 0.78% respectively (Table 2). These elements might have entered during the preparatory process of Abhraka Bhasma due to the crucibles used or as minor byproducts of reactions taking place. Oxygen, Silicon, Potassium and iron are found to be occupying greater weight % in the sample. However, when peaks omitted were only in the 8 keV range, Potassium was found to be in lesser amount than previously recorded (Table 3). Oxygen was slightly on the higher side while Iron and Potassium were on slightly lower side when compared to the previously readings (Table 2).

Bhasmas are tiny enough, having nano size, to work into blood circulation imparting pharmacological efficacy to the Bhasma. The commercial Abhraka Bhasma sample analyzed specified as the criterion for the final product conforming to all the traditional parameters under Bhasma *pariksha* (examination of properly prepared Bhasma). This can be one of the important factors for standardization of Bhasmas. Nanosize also allows the phenomenon of *rekhapurna* and *varitara* to develop.[12] Reduction in particle size facilitates absorption and assimilation of the Bhasma in the system.[13] The particle size recorded can be characterized as the desired specification of the test drug. Nano size particle of the sample may be attributed to the grinding of raw materials for a long duration (similar to the Top-Down approach of the formation of nanostructure materials in modern nanotechnology) as well as the heat treatment which causes the change in the chemical nature of the raw materials.[14] These nanosized-particles constituting Abhraka Bhasma possibly impart the therapeutic properties to it.

Recent studies have claimed that the herbo-mineral formulations of Ayurveda constituting Bhasmas to be equivalent and in tune with nanotechnology.[15] The present study proved this. This physico-chemical characterization study will help in a reappraisal of Bhasma preparations (preparations, where herbs, minerals and metals are incinerated to ash under supervised conditions) as novel nano-technological applications giving a new thrust to Ayurvedic pharmaceutics.

Our modern analytical techniques of EDXRF, FEG-SEM and EDS confirmed the absence of organic compounds and mercury in the commercial Abhraka Bhasma sample characterized in this study. The absence of mercury in the present Bhasma preparation indicates proper incineration yielding a high-quality product with better potential acceptability.[16] It puts to rest concerns about presence of heavy metals and organic compounds in Ayurvedic preparations, which otherwise clouds popular use of Ayurvedic medicines abroad.

The thorough physico-chemical characterization of the Abhraka Bhasma could be of some help to establish the probable mechanisms involved in the conversion of the metals into the Bhasmas. This characterization study establishes that incineration has been complete and the mineral is in its Bhasma form. This study reveals that Abhraka Bhasma particles are in their oxide state.[17]

This work is a very intricate area of research as the processes involved in actions of medicines on human bodies must depends on number of parameters, not simply the chemical reactions.[18] A well planned long term research can possibly enrich our understanding of the wonder drug, Abhraka Bhasma or Bhasma in general. Although, these experimental observations will help in the quality assurance and standardization of Bhasma yet some more work is needed in this direction to find out more simplified methods which can be used for routine testing in the Ayurvedic industry. Inclusion of analytical techniques becomes a necessary prerequisite to evaluate the quality of Bhasma preparations and formulations.

CONCLUSION

In conclusion, the Aryuvedic formulation, Abhraka Bhasma, analysed satisfies the criteria mentioned in the Ayurvedic Pharmacopoeia. The state-of-the-art techniques of EDXRF and high-resolution FEG- SEM imaging combined with low-voltage EDS analyses are very appropriate tools for reliable analyses of the structural and chemical composition of Abhraka Bhasma. The results obtained show that the sample doesnot contain any toxic metal even in trace levels and is of standard quality. The data of the present study suggests that the characterized Abhraka Bhasma sample, constituting nanoparticles, is in tune with nanotechnology of contemporary era.

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REFERENCES

1. Kapoor RC, Some observations on the metal-based preparations in the Indian Systems of Medicine, Ind. J. Trad. Knowledge, 9(3), 2010, 562-575.
2. http://healthstoreindia.com/buy/abhrak-bhasma/
3. Mohaptra S, Jha CB, Physicochemical characterization of Ayurvedic bhasma (*Swarna makshika bhasma*): An approach to standardization, Int. J. Ayurveda Res., 1(2), 2010, 82–86.
4. http://pharmaquest.weebly.com/uploads/9/9/4/2/9942916/factor_for_drug_absorption.pdf
5. Jayakars M. Evaluation of Krimihara property (Antihelminthic Effect) of Abhraka Bhasma prepared with Kumari Swarasa - an experimental study[dissertation]. Bangalore, Rajiv Gandhi University, 2011, 117.
6. Gupta KL, Virupaksha, Neeraj K, Characterization of *Tarakeshwara Rasa*: an Ayurvedic herbomineral formulation, Ayu., 33(3), 2012, 406–411.
7. Dubey N, Dubey N, Mehta RS, Saluja AK, Jain DK, Physicochemical and pharmacological assessment of a traditional biomedicine: *Mukta shouktic* Bhasma, Songklanakarin J. Sci. Technol., 31 (5), 2009, 501-510.
8. Rasheed A, Anvesh M, Madhu NM, Standardization of Bhasma-importance and prospects, J. Pharm . Res., 4(6), 2011, 1931.
9. Singh SK, Gautam DNS, Rai SB, Synthesis, Characterization and histopathological study of a lead-based Indian traditional drug: *Naga Bhasma,* Indian J. Pharm. Sci., 72(1), 2010, 24–30.
10. Kapoor RC, Some observations on the metal-based preparations in the Indian Systems of Medicine, Ind. J. Trad. Knowledge, 9(3), 2010, 562-575.
11. Horie M, Kato H, Fujita K, Endoh S, Iwahashi H, In vitro evaluation of cellular response induced by manufactured nanoparticles, Chem. Res. Toxicol., 25(3), 2012, 605-19.
12. Mohaptra S, Jha CB, Physicochemical characterization of Ayurvedic bhasma (*Swarna makshika bhasma*): An approach to standardization, Int . J. Ayurveda Res., 1(2), 2010, 82–86.
13. Mulik SB, Jha CB, Physicochemical characterization of an Iron based Indian traditional medicine: *Mandura Bhasma,* Anc. Sci. Life., 31(2), 2011, 52–57.
14. Singh SK, Gautam DNS, Rai SB,Synthesis, Characterization and Histopathological Study of a Lead-Based Indian Traditional Drug: *Naga Bhasma,* Indian J. Pharm. Sci., 72(1), 2010, 24–30.
15. Virupaksha GKL, Pallavi G, Patgiri BJ, Kodlady N, Relavance of Rasa Shastra in 21st century with special reference to lifestyle disorders (LSDs), IJRAP, 2(6), 2011, 1628-1632.
16. Brown CL, Bushell G, Whitehouse MW, Agrawal DS, Nanogoldpharmaceutics-Characterization of the gold in Swarna bhasma, a microparticulate used in traditional Indian medicine, Gold Bulletin, 40(3), 2007, 245-250.
17. Sarkar PK, Chaudhary AK, Ayurvedic Bhasma: the most ancient application of nanomedicine, J. Scien. Industrial Res., 69, 2010, 901-905.
18. Tripathi A, Joshi B, Singh HS, Rathore JS, Sharma G, Chemical phases of some of the Ayurvedic heamatinic medicines, Int. J. Engineering Sci. Tech., 2(8), 2010, 25-32.

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