



## Pharmaceutical Standardisation

# Standard manufacturing procedure of *Tamra Bhasma*

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### Abstract

*Tamra Bhasma* (incinerated copper) is one of the main weapons in the archery of Ayurvedic practitioners. Though several methods of preparation of *Tamra Bhasma* (TB) are found in *Rasashastra* classics, several difficulties occur during the preparation of a good-quality *Bhasma*. In this study, TB was prepared and analyzed to develop the standard manufacturing procedure. Each unit operative procedure was considered as an independent processing and an attempt was made to validate each procedure. Wire used for the purpose of electrical earthing was taken for the preparation of *Bhasma*. Procedures of *Shodhana*, *Marana*, and *Amritikarana* were followed as per the classical references. Specific temperature pattern was adopted for *Putas* in the electrical muffle furnace. From 500 g of *Tamra*, 483.4 g of black colored TB was obtained after subjecting to three *Putas*. Final product was detected to be cupric sulfide in X-ray diffraction. In particle size distribution analysis 10% of the material was below the size of 2  $\mu$ m, while in inductive coupled plasma - atomic absorption spectrometry 58.56 wt% copper and 22.48 wt% of sulfur were found present in the final product along with the elements such as arsenic, lead, zinc, mercury, and manganese in traces.

**Key words:** *Bhasma*, *Putas*, standard manufacturing procedure, *Tamra*, X-ray diffraction

### Introduction

Present day demand of Ayurvedic formulations has been raised globally due to increased response toward Ayurvedic system of medicine. Hence, commercialization of Ayurvedic drug manufacturing has taken place. Therefore, certain things like standardization, quality control, and safety became essential requirements for Ayurvedic formulations. It is the need of the time to present, understand, and implement these things into Ayurvedic formulations more accurately for globalization of Ayurveda. Thus, the production of standard, effective, genuine, safe drugs of utmost quality should be given prime importance by the processing units of Ayurvedic drugs.<sup>[1]</sup> Unfortunately, Ayurvedic formulations prepared in above-said terms are fewer. This may be because of the scarcity of standard parameters to compare a prepared drug.

Metals and minerals, as such in an elemental form, cannot be used for the therapeutic purpose, since many of them are toxic to the human body.<sup>[2]</sup> But the *Rasashastriya* pharmaceutical processes like *Shodhana* (purification/

detoxification), *Marana* (incineration/calcination), etc., make them into such form (compound) that they are highly effective without any untoward effects in the therapeutic dose.<sup>[3]</sup> *Tamra Bhasma* (incinerated copper) is important amongst the metallic *Bhasmas* used for the treatment of many diseases. Though numbers of methods of *Tamra Bhasma* (TB) preparation are described in *Rasa* literature; its preparation has always been a practical problem. Moreover, improperly prepared (*Apakwa*) TB has been quoted as poison or more than a poison, because of its hazardous effects on the body.<sup>[4]</sup> To indicate its toxic potential, *Ayurveda Prakasha* have quoted *Ashtamahadoshas* (eight major ill effects).<sup>[5]</sup> Therefore, it is extremely important to prepare TB of good quality by following the classical procedures, which include *Shodhana*, *Marana*, and *Amritikarana*. This particular work is the modest attempt in the direction of development and establishment of Standard Manufacturing Procedure (SMP) for TB. For validation, each unit operative procedure was considered as an independent processing. Final product (TB) was analyzed by classical tests, physicochemical parameters, and by applying some advanced analytical techniques.

### Materials and Methods

*Tamra* wire of 0.5 mm diameter was collected from an electrician. There was no coating over the wire. This wire was converted to thin sheet of 36 gauge thickness by passing it through a roller press. The sheet was then converted to pieces

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of 1 × 1 cm. A brief description of the materials used during the preparation of TB is given in Table 1.

*Tila Taila* (sesame oil), *Parada* (mercury), and *Gandhaka* (sulfur) were collected from Pharmacy, Gujarat Ayurved University, Jamnagar. *Kulattha* (*Dolichos biflorus* Linn.) seeds, *Surana Kanda* (corm of *Amorphophyllus campanulatus* Linn.) and *Nimbu* (*Citrus medica* Watt.) were procured from the local market of Jamnagar. All the herbal materials were identified and authenticated in the Pharmacognosy Laboratory of Institute for Post Graduate Teaching and Research in Ayurveda, Gujarat Ayurved University, Jamnagar. *Gomutra* (cow urine) was collected from local cow shed.

### Process validation of Samanya Shodhana (general purification) of Tamra

*Samanya Shodhana* of *Tamra* was carried out by *Nirvapa* (heating and quenching).<sup>[6]</sup> *Kanji* (sour gruel), *Kulattha Kwatha* (decoction of horse gram), and *Takra* (buttermilk) were prepared as per the references of *Parada Samhita*,<sup>[7]</sup> *Sharangadhara Samhita*,<sup>[8]</sup> and *Sushruta Samhita*<sup>[9]</sup> respectively. The 500 g of *Tamra* (pieces of 1 × 1 cm and 36 gauge thickness) was heated in an iron ladle to red-hot stage and quenched in each liquid media for 7 times. Temperature at the time of red-hot stage was taken by a thermocouple. Each time, liquid media was taken fresh and gravimetrically equal to the *Tamra* (500 ml × 1 × 7 = 3500 ml).

### Process validation of Vishesha Shodhana (specific purification) of Tamra

*Vishesha Shodhana* of *Tamra* was carried out as per the reference of *Rasaratnasamucchaya*.<sup>[10]</sup> *Samanya Shodhita Tamra* (3 batches) was boiled in freshly collected *Gomutra* (3 L × 3 batches = 9 L) for 3 h in *Dola Yantra*. It was then washed with warm water and dried.

### Process validation of Parada Shodhana

*Parada Shodhana* was done as per the classical reference of *Rasatarangini*.<sup>[11]</sup> *Ashuddha Parada* (impure mercury): 500 g × 3 batches = 1500 g, *Sudha Raja* (lime powder): 500 g × 3 batches = 1500 g, *Nistusha Lashuna* (garlic): 500 g × 3 batches = 1500 g, and *Saindhava Lavana* (rock salt): 250 g × 3 batches = 750 g were taken as ingredients. *Parada* was triturated with *Sudha Raja* for 3 days. On the 4<sup>th</sup> day, it was filtered through a cotton cloth and procured *Parada* was triturated with *Nistusha Lashuna* and *Saindhava Lavana* until the *Kalka* (paste) became

black. Then it was washed with hot water and filtered through a cloth, weighed and stored in a glass bottle.

### Process validation of Gandhaka Shodhana

The classical reference of *Rasatarangini* was followed throughout the procedure.<sup>[12]</sup> *Ashuddha Gandhaka*: 1 kg × 6 batches = 6 kg, *Godugdha* (cow milk): 2 L × 3 times × 6 batches = 36 L (gravimetrically double to the *Gandhaka* each time), *Goghrita* (cow ghee): 250 g × 3 times × 6 batches = 4500 g (gravimetrically 1/4<sup>th</sup> to *Gandhaka* each time) were taken for the practical work. In the reference, volume of *Godugdha* is not mentioned. Here, the volume was considered, so as to dip the melted *Gandhaka* completely. Furthermore, the amount of *Goghrita* was not taken as per the reference, because it was observed in pilot study that only 1/4<sup>th</sup> of *Goghrita* is sufficient to melt the *Gandhaka*. Powdered *Gandhaka* was heated with *Goghrita* over *Mandagni* (mild heating). After melting, it was poured into hot *Godugdha* through *Goghrita* smeared cotton cloth. A solid mass with some granular *Gandhaka* was taken out and washed with hot water. The procedure was repeated twice. After drying, it was powdered, weighed and kept in a glass bottle.

### Process validation of preparation of Kajjali

*Shuddha Parada*: 500 g × 3 batches = 1500 g, *Shuddha Gandhaka*: 500 g × 3 batches = 1500 g were taken for the preparation of *Samaguna Kajjali*. In a *Khalvayantra*, *Shodhita Parada*, and *Shuddha Gandhaka* were taken in equal quantity and triturated. It was continued until the powder became black, smooth, and lusterless.

### Process validation of Tamra Bhasma

TB was prepared as per the reference of *Rasaratnasamucchaya* by the principle of *Putra* (incineration) in an Electrical Muffle Furnace (EMF).<sup>[13]</sup> *Samaguna Kajjali* equal to the amount of *Shuddha Tamra* was taken in mortar and *Nimbu Swarasa Bhavana* (wet trituration with lemon juice) was given. When paste like consistency appeared, the mixture of *Shuddha Tamra* was added in it and triturated. After drying in shade, it was kept in *Sharava* (earthen saucer) and covered by another *Sharava* and junction was sealed by double fold of *Multany Mitty* smeared clothes. It was subjected for *Putra* in EMF. On the next day, after *Swangasheetikarana* (self-cooling) *Sharava Samputa* was removed, material was collected and triturated. In subsequent *Putra* equal amount, to that of one *Puti Bhasma*, of *Samaguna Kajjali* was added and triturated well by giving *Bhavana* of

**Table 1: Brief description of drugs used in preparation of Tamra Bhasma**

Name of drug	Latin name/English name	Purpose
<i>Tila Taila</i>	Oil of <i>Sesamum indicum</i> Linn.	<i>Samanya Shodhana</i>
<i>Takra</i>	Buttermilk	
<i>Gomutra</i>	Cow urine	
<i>Kanji</i>	Sour gruel	
<i>Kulattha Kwatha</i>	Decoction of seeds of <i>Dolichos biflorus</i> Linn.	
<i>Gomutra</i>	Cow urine	<i>Vishesha Shodhana</i>
<i>Shuddha Parada</i>	Mercury (Hg) – purified	<i>Marana</i>
<i>Shuddha Gandhaka</i>	Sulfur (S) – purified	
<i>Nimbu Swarasa</i>	Fruit juice of <i>Citrus limon</i> (Linn.) Burm. f.	<i>Bhavana</i>
<i>Surana Kanda</i>	Corm of <i>Amorphophyllus campanulatus</i> Linn.	<i>Amrutikarana</i>

sufficient quantity of *Nimbu Swarasa*. After drying, this powder was subjected to *Putra*. Total three *Putra* were given to complete the preparation of *Bhasma*. Following temperature pattern was followed: 1<sup>st</sup> *Putra* – 700°C for 20 min, 2<sup>nd</sup> *Putra* – 600°C for 25 min, 3<sup>rd</sup> *Putra* – 500°C for 30 min.

### Process validation of Amrutikarana of Tamra Bhasma

*Amrutikarana* of TB was done as per the reference of *Rasa Tarangini*.<sup>[14,15]</sup> *Surana Kanda*, *Shuddha Gandhaka*, and *Nimbu Swarasa* were used in this procedure. TB was triturated with ½ part of *Shuddha Gandhaka* in mortar and *Bhavana* of *Nimbu Swarasa* given. After proper trituration, a round bolus was prepared and dried in sunlight. *Surana* weighing 2.5 kg was cut into two halves horizontally. A round pit was made in the middle of both the halves. Dried bolus was kept in it and the two halves were joined together. A thick layer of *Kapadmitty* (mud smeared cloth) was done over it. It was then dried in sunlight. This was kept in EMF at the temperature of 500°C for 15 min. After *Swangasheetikarana* it was removed, triturated and stored in an airtight glass bottle.

### Analysis of final product

To lay down the standards, raw *Tamra*, in process material and final product were analyzed physico-chemically. Final product (TB) was analyzed by using different organoleptic parameters like *Varna* (color), *Rasa* (taste), etc.; classical tests like *Varitaratwa*, *Rekhapurnatwa*, *Apunarbhavatwa*, *Uttama/Unama*, *Niruttha*, etc.,<sup>[16]</sup> and *Dadhi Pariksha* (curd test)<sup>[17]</sup> were performed. Other parameters like *Avami*, *Niswaduta*, etc., were also applied.<sup>[18]</sup> Modern physico-chemical parameters like loss on drying, ash value, acid insoluble ash, and water soluble extractive, etc., were performed on three samples of TB.<sup>[19]</sup> Sophisticated instrumental analytical techniques like scanning electron microscopy (SEM), particle size distribution (PSD), and inductive coupled plasma - atomic absorption spectrometry (ICP-AES) of one sample of TB was carried out. Instrument used for the particle PSD was Sympatec HELOS (H1004) SUCCELL. Instrument used to perform quantitative elemental evaluation (ICP-AES) of the sample was - Inductive Coupled Plasma Spectrometer, Model: ARCOS from M/s. Spectro, Germany, R.F. Generator: Maximum of 1.6 KW, 27.12 MHz.

## Observations and Results

During the *Samanya Shodhana* of *Tamra*, all *Tamra* flakes became red-hot at 460°C. As soon *Tamra Patra* were quenched in media typical “hissing” sound was produced with boiling of

that media. After every quenching some residue in the form of powder was observed. After complete *Shodhana*, *Tamra* turned into a mixture of black coarse powder and pieces of *Tamra*. The average time taken by *Tamra* to become red and change in weight during the procedure is depicted in Table 2.

During the *Vishesha Shodhana* of *Tamra*, froth was observed on *Gomutra* after 10 min of boiling. Temperature of *Gomutra* was recorded between 96°C and 102°C throughout the procedure. After *Swedana*, *Tamra* became greenish in color. Color of *Gomutra* turned to blackish green with a strong irritating smell. The weight loss of *Tamra* after *Vishesha Shodhana* is depicted in Table 3.

In *Parada Shodhana* procedure, after trituration with *Sudha*, almost all *Parada* mixed with it after 4 h of trituration and color of the mixture turned gray. White *Lashuna Kalka* changed to black within 2 h of trituration. After this procedure, 451.5 g of *Shuddha Parada* was procured and 48.5 g (9.7%) of weight loss was observed in *Parada*.

During the *Gandhaka Shodhana* procedure, *Gandhaka* melted in 14 min (average) and the temperature recorded at its time of melting was 116°C. Crystalline dark yellow *Gandhaka* turned granular and dull yellow after *Shodhana*. Average 16.40 g (1.64%) decrease in weight was observed.

During the *Kajjali* preparation, color of the mixture started to become black after 12 h of trituration which turned dark black after 16 h trituration. *Varitaratva* (floating on stagnant water), *Rekhapurnatva* (filling the furrows when rubbed between two fingers), and *Nischandratva* (lustreless) were found after average 26.33 h of *Mardana* (trituration). On an average, 960 g of *Kajjali* was obtained and 40 g (4%) loss in weight was observed.

During the *Bhasma* preparation, cracks were observed on *Sharava* after taking out from EMF. Whitish layer of mercury was observed near the door of EMF. After first *Putra*, *Tamra Patra* became so brittle that it was getting powdered on rubbing between two fingers. Black, smooth, and very fine *Bhasma* was obtained after 3 *Putra*. Particulars of *Marana* procedure, duration of temperature given to *Putra*, specific observations and other observations regarding change in weight, etc., are depicted in Tables 4-7.

In the process of *Amrutikarana*, three hours of trituration was required for the consistency of bolus formation. After *Putapaka*, *Surana* was found totally burnt. Bolus inside was black and breakable by some pressure. After trituration it converted to black, smooth, and fine powder [Table 8]. When different samples of *Tamra* analyzed quantitatively, 99.89%, 90.84% of copper and 0.10%, 5.53% of iron was detected in raw and

**Table 2: Average time to become red hot and change in weight after Samanya Shodhana**

Media	Avg. time taken to become red hot (h:min)	Wt. of residue after 7 <i>nirvapas</i> (g)	Avg. wt. (g)	Avg. wt. ↑/↓ (g)	Avg. wt. ↑/↓ (%)
<i>Tila Taila</i>	7:46	4	514.18	14.18 ↑	1.42 ↑
<i>Takra</i>	8:53	NM*	489.47	11.53 ↓	2.30 ↓
<i>Gomutra</i>	8:28	53.8	478.38	22.62 ↓	4.52 ↓
<i>Kanji</i>	8:54	112.9	465.10	34.90 ↓	6.98 ↓
<i>Kulattha Kwatha</i>	8:59	90.4	460.20	39.80 ↓	7.38 ↓

\*Not measurable, ↑: Increase, ↓: Decrease, Wt.: Weight, Avg. wt.: Average weight

*Shuddha Tamra* respectively. TB complied with all the classical parameters [Table 9]. Physicochemical and advances analytical characters of TB are depicted in Table 10.

## Discussion

Aim of the present study was to standardize the preparation method of TB. The *Bhasmas* prepared with *Parada* or *Parada Bhasma* as media are considered superior to others.<sup>[20]</sup> In this study, *Kajjali* (black sulfide of mercury - HgS)<sup>[21]</sup> was used as media for TB preparation.

99.89% of copper was found in raw *Tamra* which indicates the high purity of raw material. Thin sheets of copper were satisfying almost all the classical parameters for *Nepalaka Tamra*.<sup>[22]</sup> Copper wire taken for the study is used for electrical earthing and that is why it was in a very pure form. Slight impurity in it decreases its electric conductivity.<sup>[23]</sup> Hence these types of wires should be used as raw material for the preparation of TB.

For *Samanya Shodhana* of *Tamra*, it was heated and after the red-hot it was quenched for 7 times in *Tila Taila*, *Takra*, *Gomutra*, *Kanji*, and *Kulattha Kwatha* in order. These are acidic,

acidic, basic, acidic and basic media in order. This specific order disrupts the internal structure of *Tamra* during the process. During *Shodhana*, color of *Tamra* became black. This is because during red-hot state, *Tamra* reacts with atmospheric oxygen and steam to form cupric oxide (CuO) which is black in color,<sup>[24]</sup> and reaction of *Tamra* occurs mainly on surface. It was observed that as *Shodhana* procedure advances *Tamra* takes comparatively more time to get complete red hot [Table 4]. Conductivity of heat is more in metallic form than compound state. So at later state of *Shodhana*, *Tamra* and its compound (CuO which is formed on the surface of *Tamra* flakes) took more time to become complete red hot.

At early stage of *Shodhana*, cracks were seen at the surface of *Tamra* flakes and finally, some coarse powder was observed. Repeated heating and cooling of *Tamra* flakes causes disruption in compression-tension equilibrium leads to cracks on the flake surface (stress corrosion theory).<sup>[25]</sup> During red hot state, compounds are formed on the surface of *Tamra* flakes. Expansibility differs from metal to compound on heating [generally expansibility of compound is less than metal (theory of thermal expansion).<sup>[26]</sup> So on repeated heating cracks are seen on the surface leading to breaking of *Tamra* flakes into coarse and some fine powder. After every seven *Nirvapas* this powder was found as sediment in media. Maximum amount of sediment (average 112.9 g) was found in *Kanji* medium [Table 3]. *Kanji* falls under the strong acidic category and also it has *Tikshna* property.<sup>[27]</sup> In *Takra* medium sediment was not observed but black particles were seen suspended in it, which was difficult to collect. In *Tila Taila* media, 1.42% weight gain was observed in spite of weight loss after *Nirvapa* process. This may be because of the adhered *Tila*

**Table 3: Average weight loss after Vishesha Shodhana of Tamra**

Avg. wt. of Samanya Shodhita Tamra (g)	Avg. amount of Gomutra required (ml)	Avg. wt. loss (g)	Avg. wt. loss (%)
450	3000	2.3	1.96

Avg. wt.: Average weight

**Table 4: Particulars of Tamra Marana**

Batch no.	Materials	Quantity		
		1 <sup>st</sup> Puta	2 <sup>nd</sup> Puta	3 <sup>rd</sup> Puta
1	<i>Shuddha Tamra</i>	450.0 g	1 <sup>st</sup> Puta Marita Tamra	2 <sup>nd</sup> Puta Marita Tamra
	<i>Samaguna</i>		465 g	472.5 g
	<i>Kajjali</i>	450.0 g	465 g	472.5 g
	<i>Nimbu Swarasa</i>	200 ml	330 ml	335 ml
2	<i>Shuddha Tamra</i>	440.0 g	1 <sup>st</sup> Puta Marita Tamra	2 <sup>nd</sup> Puta Marita Tamra
	<i>Samaguna</i>		458 g	467.8 g
	<i>Kajjali</i>	440.0 g	458 g	467.8 g
	<i>Nimbu Swarasa</i>	200 ml	320 ml	330 ml
3	<i>Shuddha Tamra</i>	455.0 g	1 <sup>st</sup> Puta Marita Tamra	2 <sup>nd</sup> Puta Marita Tamra
	<i>Samaguna</i>		466 g	480.3 g
	<i>Kajjali</i>	455.0 g	455.0 g	480.3 g
	<i>Nimbu Swarasa</i>	200 ml	320 ml	340 ml

**Table 5: Duration and temperature given for Putas**

Put a no.	Maximum temp. given in EMF (°C)	Time taken to reach that temp. (h:min)	Duration of maximum temp. (min)	Total duration of giving temp. (h:min)	Time taken for Swangasheetikarana (h:min)
1	700	2:5	20	3:0	20:0
2	600	2:1	25	2:25	18:0
3	500	2:0	30	2:20	15:0

EMF: Electric muffle furnace, temp.: Temperature

*Taila*; which could not be removed even after thorough wash with hot water.

Out of six liquid media used for *Shodhana* procedure, three were acidic (pH ranging from 3 to 3.5) and others were weak bases (pH ranging from 7.3 to 8.9). The alternate heating and quenching in these media lead to corrosive changes in the metal and may also cause removal of acid and alkali soluble impurities from the metal. It should also be noted that these media were among the naturally and easily prepared source of acid and base at the ancient times.

*Vishesha Shodhana* of *Tamra* is a simple process of *Swedana* in *Dola Yantra*, specific to *Tamra*. Possible hypothesis for conduction of this process may be further micropurification and impregnation of qualities for *Bhasma* preparation. In this process, components of *Gomutra* may pierce through the micropores and cracks created during *Samanya Shodhana* and may produce the required change specific to *Tamra* for further process. After *Vishesha Shodhana*, observed weight loss was may

be due to the reaction between ammonia from *Gomutra* and copper to form the cuprammonium ion  $[Cu(NH_3)_4]$ , a chemical complex which is water-soluble.<sup>[28]</sup> This complex gets washed away during washing with hot water.

Ingredients used in the *Parada Shodhana* contained *Kshariya* (alkaline) materials like *Sudha* (lime) and *Saindhava* (rock salt). They may be removing the alkaline soluble impurities along with trace elements present in *Parada*. It was observed that when *Parada* was triturated with *Sudha*, it was converted to powder form, which may be referred as Grey powder (calomel). It is difficult to procure the whole amount of *Parada* by *Vastra Galana* (straining through cloth) process as mentioned in classics. So it was washed with hot water. During this procedure loss of *Parada* with water should be checked.

For the process of *Gandhaka Shodhana*, powdered sulfur was taken for the sake of easy melting. *Mandagni* was given to avoid burning of sulfur. Cloth was smeared with *Ghee* to avoid sticking of *Gandhaka* to the cloth. Hot *Godugdha* was taken for pouring of melted *Gandhaka* since it facilitates the granule formation while cold milk forms hard mass of *Gandhaka*. After each *Dhalana* (melting and pouring), *Gandhaka* was thoroughly washed with hot water to remove fat contents of milk and *Ghee*.

Loss in weight of *Kajjali* was observed because of dusting of mixture during trituration and some of it remained adhered to *Khalva* (mortar) which was difficult to collect.

Due to unavailability of *Jambira* (*Citrus limon* Linn. Burm.f.) in Jamnagar region, *Nimbu* (*Citrus medica* Watt.) was used. EMF was used for *Puta* to avoid variations such as weight and quality of cow dung cakes, seasonal variations that occur in classical *Puta*. EMF is a modified instrument by which a desired highest temperature and duration of heat can be adjusted; chance of heat loss is less and a homogeneous temperature pattern can be provided. Nowadays, many of the Ayurvedic pharmacies are using this instrument for preparing *Bhasmas*. Therefore it was preferred for *Putapaka* to provide a standard heating pattern.

*Chakrikas* were not prepared. In pilot study, it was observed that when *Chakrikas* were prepared, they became hard after every *Puta* and took nine *Putas* to form *Bhasma*. However, in a batch where *Chakrikas* were not prepared and only mixture was layered; it took only four *Putas* to form *Bhasma*. Thickness of layered mixture was around 1 cm. Although, thickness of the layer (1 cm) was more than that of traditional *Chakrika* (0.5 cm), which may cause less heat flow through the layered mixture according to Fourier's law.<sup>[29]</sup> However, in layered method also temperature was given for more time to complete the chemical reaction and to form a desired compound.

**Table 6: Specific observations and results of Tamra Bhasma preparation after each Puta**

Batch no.	Putra no.	Wt. of Bhasma (g)	Color of Bhasma	Taste	Curd test
1	1	465.0	Blackish brown	Strongly metallic	-ve
	2	472.5	Blackish	Slightly nauseating	-ve
	3	476.3	Black	Tasteless	+ve
2	1	458.0	Blackish	Strongly metallic	-ve
	2	467.8	Blackish	Slightly astringent	-ve
	3	470.7	Black	Taste less	+ve
3	1	455.0	Blackish brown	Strongly metallic	-ve
	2	480.3	Black	Tasteless	-ve
	3	482.6	Black	Tasteless	+ve

Wt.:Weight

**Table 7: Increase in weight of Tamra Bhasma**

Batch no.	Weight (g)		Weight increased (g)	Weight increased (%)
	Initial	Final		
1	450	476.3	26.3	5.8
2	440	470.7	30.7	6.9
3	455	482.6	27.6	6.06
Average	448.33	476.53	28.2	6.25

**Table 8: Observations during Amrutikarana of Tamra Bhasma**

Batch no.	TB taken for Amrutikarana (g)	Surana kanda (g)	Shuddha gandhaka (g)	Nimbu swarasa (ml)	TB after amrutikarana (g)	Wt. gain (g)	Wt. gain (%)
1	470	2550	235	230	483.6	13.6	2.9
2	465	2730	232.5	210	480.8	15.8	3.4
3	475	2390	237.5	210	485.7	10.7	2.3
Avg.	470	2556.7	235	216.7	483.4	13.4	2.9

Wt.:Weight;Avg.:Average;TB: Tamra Bhasma

For the first *Putra*, *Nimbu Swarasa* was required gravimetrically 1/4<sup>th</sup> of the total amount of *Shuddha Tamra* and *Samaguna Kajjali*. This amount of *Nimbu Swarasa* increased from second *Putra* onwards. This is because of reduced particle size and increased surface area of the material.

In *Rasamrutam*, Yadavaji has told that higher temperature in initial *Putas* and lower temperature in later should be given during preparation of TB.<sup>[30]</sup> Hence, EMF was selected for the *Putas* to regulate the temperature owing to its convenience. First, *Putra* was given 700°C and maximum temperature of *Putas* was reduced to 600°C and 500°C in later *Putas* [Table 5]. This particular temperature pattern was followed as a result of a pilot study for preparation of TB, which was carried out before going through this study.

The color of TB is black (*Krishna*).<sup>[31]</sup> In the process of TB preparation, copper is converted to its sulfide form in major since the sulfur (dissociation product of HgS) is an accompaniment to the metal in the processing. During the heat treatment for multiple times (*Putra*), some sulfides may get converted to oxide.

**Table 9: Organoleptic and other classical characters of Tamra Bhasma**

Parameters	Tamra Bhasma
<i>Shabda</i> (sound)	No sound when rubbed between fingers
<i>Sparsha</i> (touch)	<i>Mridu</i> (smooth)
<i>Rupa</i> (color/appearance)	<i>Krishna</i> (black), powder
<i>Rasa</i> (taste)	<i>Niswadu</i> (tasteless), <i>Awami</i> (non-nauseating/vomiting)
<i>Gandha</i> (odor)	Not specific
<i>Varitaratva</i> (floating on water)	+ve
<i>Rekhapurnatva</i> (filling the furrows when rubbed between two fingers)	+ve
<i>Nishchandravta</i> (lusterless)	+ve
<i>Dantagre Kachkachaabhava</i> (no grating sensation when chewed between teeth)	+ve
<i>Dadhi Pariksha</i> (curd test)	Passed

+ve: Compiled

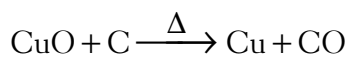
**Table 10: Physicochemical and advanced analytical characters of Tamra Bhasma**

Parameters	Tamra Bhasma
Loss on drying (%w/w)	0.13
Ash value (%w/w)	98.10
Acid insoluble ash (%w/w)	2.32
Water soluble extract (%w/w)	0.78
CS <sub>2</sub> soluble extract (%w/w)	1.33
Phase identification (XRD)	CuS
Particle size distribution (VMD)	28.70 μm
Element content (ICP-AES) wt%	Cu: 58.56 S: 22.48, Fe: 0.31 As, Pb, Hg, Mn, Zn: Traces Cd, Se: Not detected

XRD: X-ray diffraction; Wt.: Weight; VMD: Volumetric mean diameter, ICP-AES: Inductive coupled plasma-atomic emission spectrometry, CuS: Cupric sulfide

Because metallic sulfides when heated in air get converted to oxide of the metal and sulfur dioxide.<sup>[32]</sup> Therefore, TB may be considered as a mixture of copper oxides (Cu<sub>2</sub>O, CuO), copper sulfides (Cu<sub>2</sub>S, CuS) and other trace elements. CuO is brown in color,<sup>[33]</sup> hence after first two *Putas* color of *Bhasma* was brownish black. It can be said that it was a combination of all these compounds. When *Bhasma* was properly formed, it was black in color. CuO and cupric sulfide (CuS) are black in color.<sup>[31]</sup> Therefore, it can be inferred that TB is either CuO or cuprous sulfide or combination of both. Average 476.53 g of TB was obtained from 448.33 g of *Shuddha Tamra*. 6.25% increase in weight after *Marana* may be attributed to formation of the oxides and sulfides of copper as mentioned earlier. Inorganic contents of *Nimbu Swarasa* (9.45% w/w) also lead to increase in weight of TB.

For the *Amrutikarana* 470 g of TB *Surana Kanda* of 2.5 kg was required. Same temperature was applied at which the *Bhasma* was formed. 2.9% increase in weight after *Amrutikarana* may be attributed to inorganic contents (mainly calcium oxalate crystals) of *Surana Kanda*. Organic contents of *Surana* act as a source of carbon. An unstable metallic compound (especially oxides) can be reduced to metallic state during this procedure by the carbon reduction process.<sup>[34]</sup>



This metallic copper can be further reduced to sulfide in presence of sulfur. However, as metallic compound should not be changed on the particular temperature in which it is formed. Hence it can be inferred that the process of *Amrutikarana* removes any unstable compound (CuO in this case) and makes the product more stable (CuS).

The color of TB obtained was black [Figure 1]. Color indicates the formation of specific compound. Here black color indicates the formation of either CuO or CuS of copper or both. Tests like *Rekhapurnatva* and *Varitaratva* indicate the lightness and fineness of the *Bhasma*. *Awami* nature of TB indicates that there is no free copper or any unwanted compound (like copper sulfate) in it. Further no discoloration in curd test even after 48 h proves that there is no free copper or copper sulfate in final product. *Awami* and no discoloration in curd test should be considered as main tests to assess the properly prepared TB.



**Figure 1: Appearance of final product (Tamra Bhasma)**

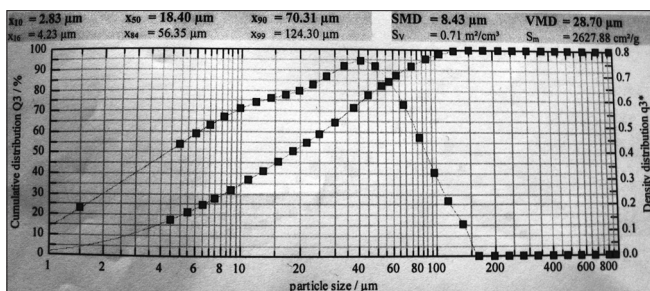


Figure 2: Particle size distribution of Tamra Bhasma

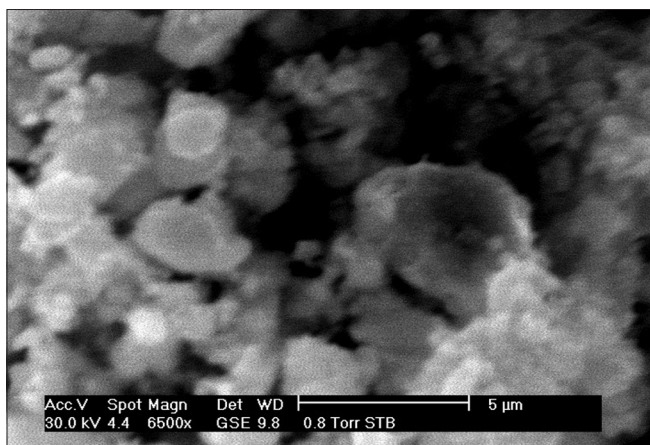


Figure 4: SEM images of Tamra Bhasma (at resolution 5 μm and magnification 6500x)

In PSD analysis 10% of the material was found below 2.83 μm where as maximum material was below 124.30 μm [Figure 2]. Small particle size enhances the absorption,<sup>[35]</sup> hence the bioavailability and thus potency of the drug increases resulting in decrease in its dose. This eventually leads to decrease in dose-related side effects of the drug.

Bhasma analyzed under scanning electron microscope showed particle size less than 2 μm [Figures 3 and 4]. In X ray diffraction [Figure 5] prominent peaks of CuS were seen, which confirms that final product is sulfide form of copper. Presence of oxides cannot be denied because other peaks were also present. Furthermore, in ICP-AES 58.56 wt% of copper against 22.48 wt% of sulfur was present. As the procedure of Tamra Shodhana was carried out in an iron pan 0.31 wt% of iron was traced in Bhasma. Heavy metals like cadmium, selenium were not detected while others like arsenic, lead and mercury were present in traces. Hence TB can be considered as blend of micronutrients.

### Conclusion

The adopted method for preparation of TB can be considered as easy, convenient and SMP. The temperature pattern adopted in EMF to prepare TB in this study can be considered as a standard heating pattern. TB should be considered as combination of copper and sulfur in CuS form though presence of oxides cannot be denied. Curd test is the simple household test to finalize the preparation of TB. After three Puta and Amritakarana, particle size of TB reaches up to micron level. In TB, along with copper and sulfur as main elements other

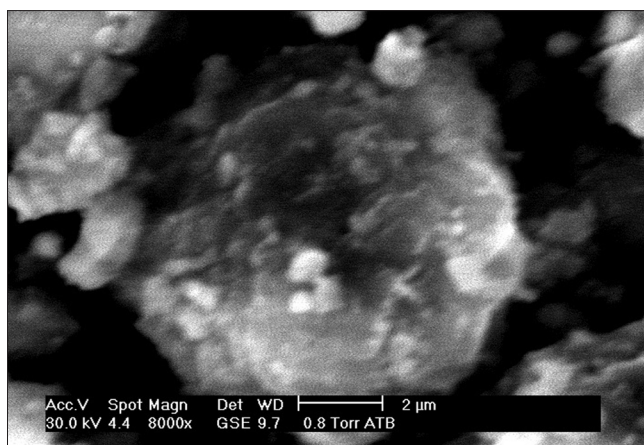


Figure 3: SEM image of Tamra Bhasma (at resolution 2 μm and magnification 8000x)

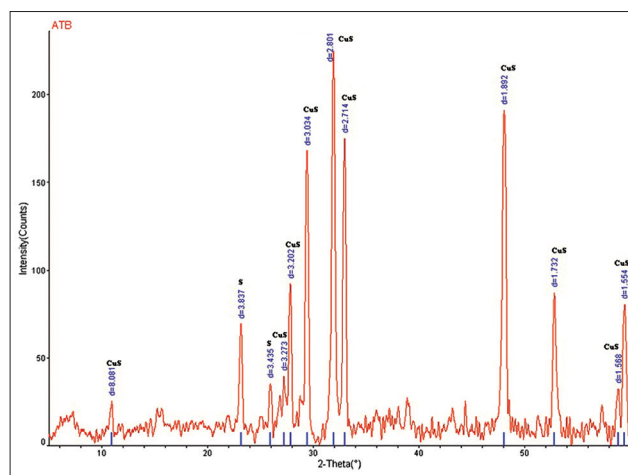


Figure 5: XRD analysis of Tamra Bhasma

elements like manganese, zinc, lead, and arsenic are also present in traces.

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## हिन्दी सारांश

### ताम्र भस्म की मानक प्रमाणित विधि

चंद्रशेखर वाय. जगताप, प्रदीपकुमार प्रजापति, बिस्वाज्योति पटगिरी, विनय जे. शुक्ला

ताम्र भस्म आयुर्वेदीय चिकित्सकों के लिये महत्वपूर्ण योग है। रसशास्त्रीय ग्रंथों में ताम्र भस्म बनाने की अनेक विधियाँ दी गयी हैं। परंतु अच्छी गुणवत्ता की भस्म बनाने में अनेक कठिनाइयाँ आती हैं। इस अध्ययन में ताम्र भस्म बनाकर उसके मानक प्रमाणित विधि के लिये विश्लेषण किया गया। प्रत्येक प्रक्रिया को एक स्वतंत्र विधि मानकर उनका विधिमान्यकरण करने का प्रयत्न किया गया। विद्युतिकरण में अर्थिन्ग के उपयोग में आनेवाली ताम्र की तार का भस्म बनाने में प्रयोग किया गया। ग्रन्थोक्त विधि से शोधन, मारण और अमृतीकरण कराये गये। पुट के लिये विद्युत ध्राष्ट्री में विशिष्ट तापक्रम रखा गया। तीन पुटों के बाद ५०० ग्रा. कच्चे ताम्र से ४८३.४ ग्रा. कृष्णवर्णी ताम्र भस्म मिला। अंतिम उत्पाद सूक्ष्म, मुलायम चूर्ण था जो XRD में CuS पाया गया। कणों के आकारमान विश्लेषण में १०% उत्पाद का आकारमान २ micron से कम पाया गया। AES ICP में ५८.५६% ताम्र एवं २२.४८% गंधक के साथ साथ अन्य मूलतत्व As, Pb, Zn, Hg, Mn अत्यल्प मात्रा में पाये गये।