


**Research Article**
**SYNTHESIS, CHARACTERIZATION AND ANTIMICROBIAL STUDY OF *VANGA BHASMA* PREPARED WITH SPECIAL REFERENCE TO *RASATARANGINI***
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**ABSTRACT**

*Vanga Bhasma* which has been prepared with *Parada* and *Haratala* or even without *Parada* and *Haratala* is widely used for a broad spectrum of diseases. It is also said to possess *Jantughna Prabhava* (Antimicrobial activity). Hence it was decided to synthesize, analyse and study the antimicrobial activity of *Vanga Bhasma* prepared with special reference to *Rasatarangini* 18/25-28. The present study deals with the preparation of *Vanga Bhasma* with reference to *Rasatarangini* 18/25-28. The synthesized *Bhasma* samples were characterized by various analytical techniques. The antimicrobial effects of these *Bhasma* samples were studied. The samples were characterized with the contemporary parameters like XRD, SEM and EDX to find out the nature of the *Vanga Bhasma* samples. These samples were further tested against certain Gram +ve, Gram-ve and fungal organisms, so as to find out the anti-microbial efficacy of the *Vanga Bhasma* samples. The adopted method for preparation of *Vanga Bhasma* (Ref. *Rasatarangini* 18/25-28) was able to produce a *Bhasma* compatible to organoleptic parameters mentioned in the ancient texts. Formation of the small sized particles as small as a nano-particle was confirmed by SEM study. Average 14 *Putas* are required to prepare *Vanga Bhasma* and XRD study confirms that Tin oxide is the major compound. *Vanga Bhasma* showed antimicrobial activity in inhibiting the growth of *Staphylococcus aureus*, *Bacillus subtilis*, *Klebsiella pneumonia*, *Escherichia coli* and *Candida albicans* with a concentration of 100mg/ml. This outcome further supports the *Krumighna* and *Jantughna* properties (anti-microbial activity) of *Vanga Bhasma*.

**KEYWORDS:** Analytical, Anti-microbial, *Maran*, *Shodhan*, *Vanga bhasma*, *Rasatarangini*.

**INTRODUCTION**

*Bhasma*, a unique dosage form, mentioned in the ancient Ayurveda texts is an incinerated metal or mineral prepared after certain rounds of processing like *Shodhana*, *Marana*, *Amritikarana* etc. The *Bhasmas* are known to offer miraculous results in clinical practice<sup>[1]</sup>. Quality of a drug depends upon its raw material selection, processing and applications with standard guidelines. It is essential to apply certain standards for manufacture of drugs so that the genuineness of the drug is not compromised. There have been concerns regarding the safety and efficacy of Ayurvedic drugs mainly the *Bhasma* with special reference to the heavy metal toxicity<sup>[2]</sup>. Keeping this fact in mind, the *Vanga Bhasma*, was prepared for the present study and analyzed for quality control checks, on the parameters described in the Ayurvedic texts as well as modern technology like SEM, EDX and XRD. These *Bhasmas* should exhibit efficacy against the disease inducing microbes. Hence, it was tested against certain Gram +ve, Gram-ve and fungal organisms, so as to find out

the anti-microbial efficacy of the *Vanga Bhasma* samples.

**MATERIALS AND METHODS**

The different materials used for the preparation of *Vanga Bhasma*; raw *Vanga* (Tin)<sup>[3]</sup>, *Parad*<sup>[4]</sup>, *Haratal*<sup>[5]</sup>, *Tila*<sup>[6]</sup> *Taila* (Sesame oil), *Takra*<sup>[7]</sup> (Butter milk), *Gomutra*<sup>[8]</sup> (Cow's urine), *Rasona*<sup>[9]</sup>, powder of *Ashwattha*<sup>[10]</sup> *Twak* (*Ficus religiosa*), *Rajika*<sup>[11]</sup>, *Saindhava*<sup>[12]</sup>, *Tandula*<sup>[13]</sup>, *Vamshapatra*<sup>[14]</sup>, *Shunthi*<sup>[15]</sup>, *Hingu*<sup>[16]</sup>, *Harida*<sup>[17]</sup>, *Masha*<sup>[18]</sup> and *Jeeraka*<sup>[19]</sup> were procured from local retailers. The *Kanji*<sup>[20]</sup> (Sour gruel), *Kulattha Kwath*<sup>[21]</sup> (Decoction of *Dolichos biflorus* Linn.), *Churnodaka*<sup>[22]</sup> (Lime water) and *Arkapatra Swaras*<sup>[23]</sup> (Expressed juice of leaves of *Calotropis procera*) were prepared in the departmental laboratory. All medicinal plants used in the study were authenticated at Department of Botany. The preparation of *Vanga Bhasma* consists of steps such as *Shodhan*<sup>[24]</sup> (*Samanya* and *Vishesha*), *Bhavana*<sup>[25]</sup> and *Maran*<sup>[26]</sup>.

**Shodhan of Vanga**

Ashuddha Vanga was subjected to Samanya and Vishesh Shodhan.

The Samanya Shodhan of Vanga was done by quenching the molten Vanga subsequently into Tila Taila, Takra, Gomutra, Kanji and Kulattha Kwath 7

times each. Then this Samanya Shodhit Vanga was subjected to Vishesh Shodhan. Here Samanya Shodhit Vanga was melted and further quenched into lime water for 7 times. Each quenching was done in a fresh media.

**Table 1: Observation regarding Weight of Vanga before and after Samanya Shodhan**

Weight (g)	Batch 1	Batch 2	Batch 3
Initial	500	500	500
Final (dried weight)	437	433.15	434.31
% change in weight	12.6	13.37	13.13

**Table 2: Observation regarding Weight of Vanga before and after Vishesh Shodhan**

Weight (g)	Batch 1	Batch 2	Batch 3
Initial	432	428.15	429.31
Final (dried weight)	416.72	414.16	415.72
Weight loss(g)	15.28	13.99	13.59
% change in weight	3.53	3.26	3.16

**Preparation of Vanga Bhasma- (Batch A3, B3, C3)- Shuddha Vanga + Ashwattha Twak Churna + Shuddha Parada + Shuddha Haratala)****Table 3: Batch wise observation regarding Maran ingredients of Vanga**

Batch No.	Shuddha Vanga (g)	Shuddha Parada (1/4 <sup>th</sup> of Vanga) (g)	Total quantity of amalgam Formation (g)	Wt. decrease in %	Shuddha Hartal (g)	Total wt.=Shuddha Vanga + Shuddha Parada + Shuddha Haratala (g)
Batch A3	138.90	34.72	157.39	9.34	69.45	226.84
Batch B3	138.05	34.51	156.11	9.53	69.02	225.13
Batch C3	138.57	34.64	157.75	8.92	69.28	227.03

**Table 4: Observation Regarding Vanga Bhasma- Batch A3**

Putra	Shuddha Vanga + Shuddha Parada + Shuddha Haratala	Bhavana Drava- Arkapatra Swaras (ml)	Wt. of Chakrikas Before Puta (Dry Chakrikas-g)	Wt. of Chakrikas After Puta (g)	Cow Dung Cakes (No.)	Cow Dung Cakes (Wt.) Kg	Max. Temp. (°C)	Time reqd. to attain the Max. Temp. (minutes)	Colour of Chakrikas after Puta	Hardness/ Softness of Chakrikas	Wt. loss after Puta (g)	% Wt. loss
1 <sup>st</sup>	226.84	90	240.4	224.28	10	5	773	18	Yellowish	Hard	2.56	1.12
2 <sup>nd</sup>	224.28	90	239.48	221.8	10	5	758	20	Light yellowish	Hard	2.48	1.10
3 <sup>rd</sup>	221.8	90	237.2	219.17	10	5	690	18	Dark Greyish	Hard	2.63	1.18
4 <sup>th</sup>	219.17	80	232.65	217.95	10	5	705	15	Dark Greyish	Hard	2.79	1.27
5 <sup>th</sup>	217.95	80	231.4	214.33	10	5	680	12	Dark Greyish	Hard	3.62	1.66
6 <sup>th</sup>	214.33	80	229.18	210.34	9	4.5	664	10	Dark Greyish	Soft	3.99	1.86
7 <sup>th</sup>	210.34	80	226.87	207.79	9	4.5	622	12	Dark Greyish	Soft	2.55	1.21
8 <sup>th</sup>	207.79	80	224.02	204.46	8	4.0	596	11	Dark Greyish	Soft	3.33	1.60
9 <sup>th</sup>	204.46	80	219.02	201.34	8	4.0	542	9	Dark Greyish	Soft	3.12	1.47
10 <sup>th</sup>	201.34	70	216.76	198.6	6	3.0	498	12	Dark Greyish	Soft	2.74	1.36
11 <sup>th</sup>	198.6	70	214.06	195.39	6	3.0	475	10	Dark Greyish	Soft	3.21	1.55
12 <sup>th</sup>	195.39	60	209.62	193.57	4	2.0	320	5	Dark Greyish	Soft	3.7	1.89
13 <sup>th</sup>	193.57	60	208.13	196.85	4	2.0	312	8	Dark Greyish	Soft	3.28	1.69

**Table 5: Observation regarding Vanga Bhasma- Batch B3**

Putra	Shuddha Vanga + Shuddha Parada + Shuddha Haratala (g)	Bhavana Drava-Arkapatra Swaras (ml)	Wt. of Chakrikas Before Putra (Dry Chakrikas-g)	Wt. of Chakrikas After Putra (g)	Cow Dung Cakes (No.)	Cow Dung Cakes (Wt.) Kg	Max. Temp. (°C)	Time reqd. to attain the Max. Temp. (minutes)	Colour of Chakrikas after Putra	Hardness/Softness of Chakrikas	Wt. loss after Putra (g)	% Wt. loss
1 <sup>st</sup>	225.13	90	240.53	227.48	10	5	693	19	Yellowish	Hard	2.35	1.00
2 <sup>nd</sup>	227.48	90	246.04	230.01	10	5	762	20	Light yellowish	Hard	2.53	1.09
3 <sup>rd</sup>	230.01	90	245.76	227.24	10	5	693	16	Dark Greyish	Hard	2.77	1.20
4 <sup>th</sup>	224.24	80	242.5	224.55	10	5	625	14	Dark Greyish	Hard	2.69	1.19
5 <sup>th</sup>	224.55	80	243.44	221.96	10	5	686	12	Dark Greyish	Hard	2.59	1.15
6 <sup>th</sup>	221.96	80	240.67	218.85	9	4.5	652	10	Dark Greyish	Soft	3.11	1.40
7 <sup>th</sup>	218.85	80	235.71	216.04	9	4.5	635	12	Dark Greyish	Soft	2.81	1.28
8 <sup>th</sup>	216.04	80	232.6	213.46	8	4.0	585	10	Dark Greyish	Soft	2.58	1.19
9 <sup>th</sup>	213.46	70	231.32	210.73	8	4.0	492	10	Dark Greyish	Soft	2.73	1.28
10 <sup>th</sup>	210.73	70	226.18	207.4	8	4.0	586	8	Dark Greyish	Soft	3.33	1.58
11 <sup>th</sup>	207.4	70	224.18	205.26	6	3.0	492	8	Dark Greyish	Soft	2.14	1.03
12 <sup>th</sup>	205.26	60	222.74	201.89	6	3.0	384	5	Dark Greyish	Soft	3.37	1.64
13 <sup>th</sup>	201.89	60	216.79	199.73	4	2.0	356	4	Dark Greyish	Soft	2.16	1.07
14 <sup>th</sup>	199.73	60	214.58	197.73	4	2.0	342	4	Dark Greyish	Soft	2	1.00

**Table 6: Observation regarding Vanga Bhasma- Batch C3**

Putra	Shuddha Vanga + Shuddha Parada + Shuddha Haratala(g)	Bhavana Drava-Arkapatra Swaras (ml)	Wt. of Chakrikas Before Putra (Dry Chakrikas-g)	Wt. of Chakrikas After Putra (g)	Cow Dung Cakes (No.)	Cow Dung Cakes (Wt.) Kg	Max. Temp. (°C)	Time reqd. to attain the Max. Temp. (minutes)	Colour of Chakrikas after Putra	Hardness/Softness of Chakrikas	Wt. loss after Putra (g)	% Wt. loss
1 <sup>st</sup>	227.03	90	241.29	224.45	10	5	705	19	Yellowish	Hard	2.58	1.09
2 <sup>nd</sup>	224.45	90	239.68	221.79	10	5	735	20	Light yellowish	Hard	2.66	1.13
3 <sup>rd</sup>	221.79	90	238.24	219.28	10	5	685	18	Dark Greyish	Hard	2.51	1.08
4 <sup>th</sup>	219.28	80	235.14	216.84	10	5	624	15	Dark Greyish	Hard	2.44	1.06
5 <sup>th</sup>	216.84	80	237.18	214.45	9	4.5	658	12	Dark Greyish	Hard	2.39	1.05
6 <sup>th</sup>	214.45	80	229.65	211.78	9	4.5	660	10	Dark Greyish	Soft	2.67	1.19
7 <sup>th</sup>	211.78	80	226.66	208.66	9	4.5	632	9	Dark Greyish	Soft	3.12	1.4
8 <sup>th</sup>	208.66	80	222.52	206.19	8	4.0	583	10	Dark Greyish	Soft	2.47	1.13
9 <sup>th</sup>	206.19	70	219.81	203.48	8	4.0	550	10	Dark Greyish	Soft	2.71	1.25
10 <sup>th</sup>	203.48	70	216.37	200.41	8	4.0	480	8	Dark Greyish	Soft	3.07	1.44
11 <sup>th</sup>	200.41	70	215.10	198.72	6	3.0	456	8	Dark Greyish	Soft	1.69	0.8

12 <sup>th</sup>	198.72	60	214.18	196.66	6	3.0	352	5	Dark Greyish	Soft	2.06	0.98
13 <sup>th</sup>	196.66	60	211.62	197.66	4	2.0	334	4	Dark Greyish	Soft	2	0.96

**Table 7: Organoleptic Characters<sup>[27]</sup>**

Parameter	Vanga Bhasma No. 3
Shabda	Anupasthita
Sparsha	Soft, no coarse Particles
Rupa	Dark grayish
Susnigdhatva	Alpa snigdha
Nischandratva	No metallic luster
Rekhapurnatva	Upasthita
Varitaratva	Upasthita
Unama	Upasthita
Rasa	Tasteless
Gandha	Not specific

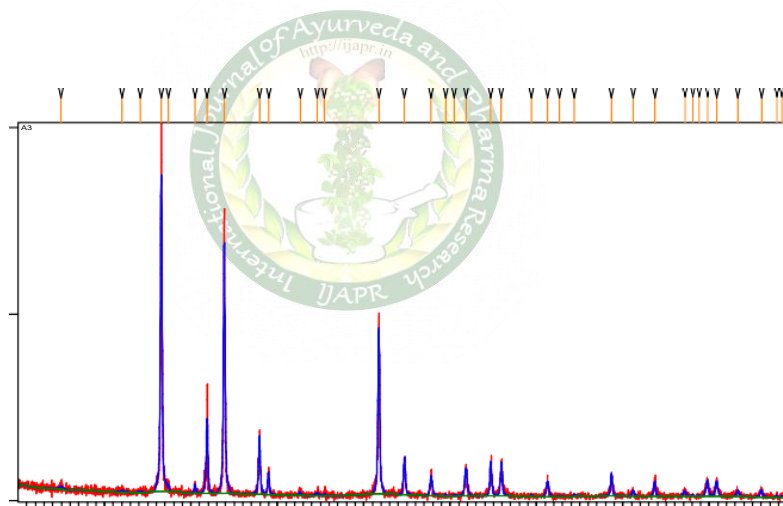
**Table 8: Analysis Details<sup>[28]</sup>**

Sr. No.	Name of Sample of Vanga Bhasma	Ash Content	Acid Insoluble Matter	Water Soluble Extractives	Alcohol Soluble Extractives	pH
1	A3	98.76%	94.43%	0.33%	0.642%	7.90
2	B3	98.18%	94.19%	0.58%	0.73%	8.75
3	C3	99.36%	93.79%	0.27%	0.72%	8.75

**Analytical Study**

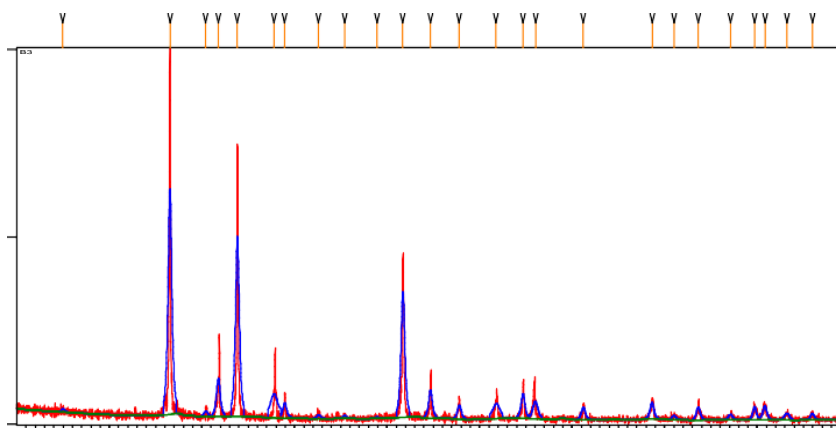
**X-Ray Diffraction Study<sup>[29]</sup>**

**Batch A3**

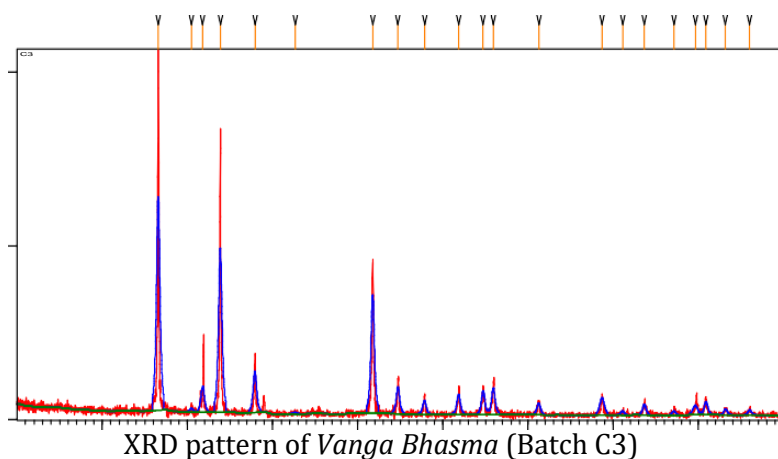


XRD pattern of Vanga Bhasma (Batch A3)

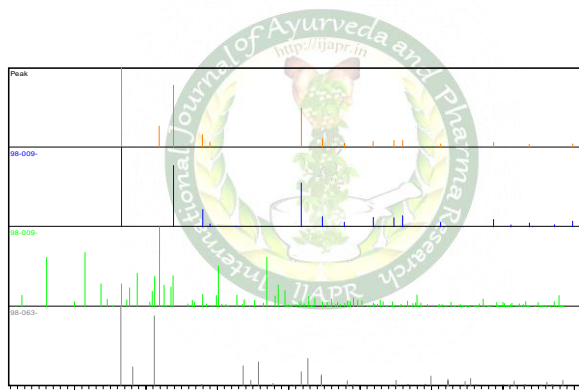
**Batch B3**



XRD pattern of Vanga Bhasma (Batch B3)

**Batch C3**XRD pattern of *Vanga Bhasma* (Batch C3)**Identified Patterns List<sup>[30]</sup>****Table 9: Showing identified pattern list of *Vanga Bhasma* (Batch A3, B3 & C3)**

Visible	Ref. Code	Score	Compound Name	Displacement [°2Th.]	Scale Factor	Chemical Formula
*	98-009-0609	87	Cassiterite	0.000	0.960	O <sub>2</sub> Sn <sub>0.912</sub>
*	98-009-7513	4	Tin(IV) Sulfide	0.000	0.439	S <sub>3</sub> Sn <sub>2</sub>
*	98-063-9165	3	Mercury Sulfide (1/1)	0.000	0.516	Hg <sub>1</sub> S <sub>1</sub>

**Plot of Identified Phases****RESULTS****Batch A3**

- Totally 16 peaks were identified in *Vanga Bhasma* (Batch A3) at different angles (2 Theta) from 26.5681 to 90.8571.
- 3 strong peaks were chosen as strong with their relative intensity and compared to standard X-ray powder diffraction file.
- 1<sup>st</sup>, 3<sup>rd</sup>, 6<sup>th</sup> peak with relative intensity of 100%, 78.24%, 49.52%, were considered as significant at 26.5681°, 33.8633°, & 51.7593° having 3.35, 2.80 & 1.76 d space value respectively.

**Batch B3**

- Totally 25 peaks were identified in *Vanga Bhasma* (Batch B1) at different angles (2 Theta) from 14.9732° to 95.9566°.
- 3 strong peaks were chosen as strong with their relative intensity and compared to standard X-ray powder diffraction file.
- 2<sup>nd</sup>, 5<sup>th</sup>, & 11<sup>th</sup> peak with relative intensity of 100%, 80.17% & 55.93% were considered as significant at 26.5469°, 33.8378° & 51.7206°, having 3.35, 2.64 & 1.76 d space value respectively.

**Batch C3**

- Totally 21 peaks were identified in *Vanga Bhasma* (Batch B1) at different angles (2 Theta) from 26.5725° to 95.9920°.
- 3 strong peaks were chosen as strong with their relative intensity and compared to standard X-ray powder diffraction file.



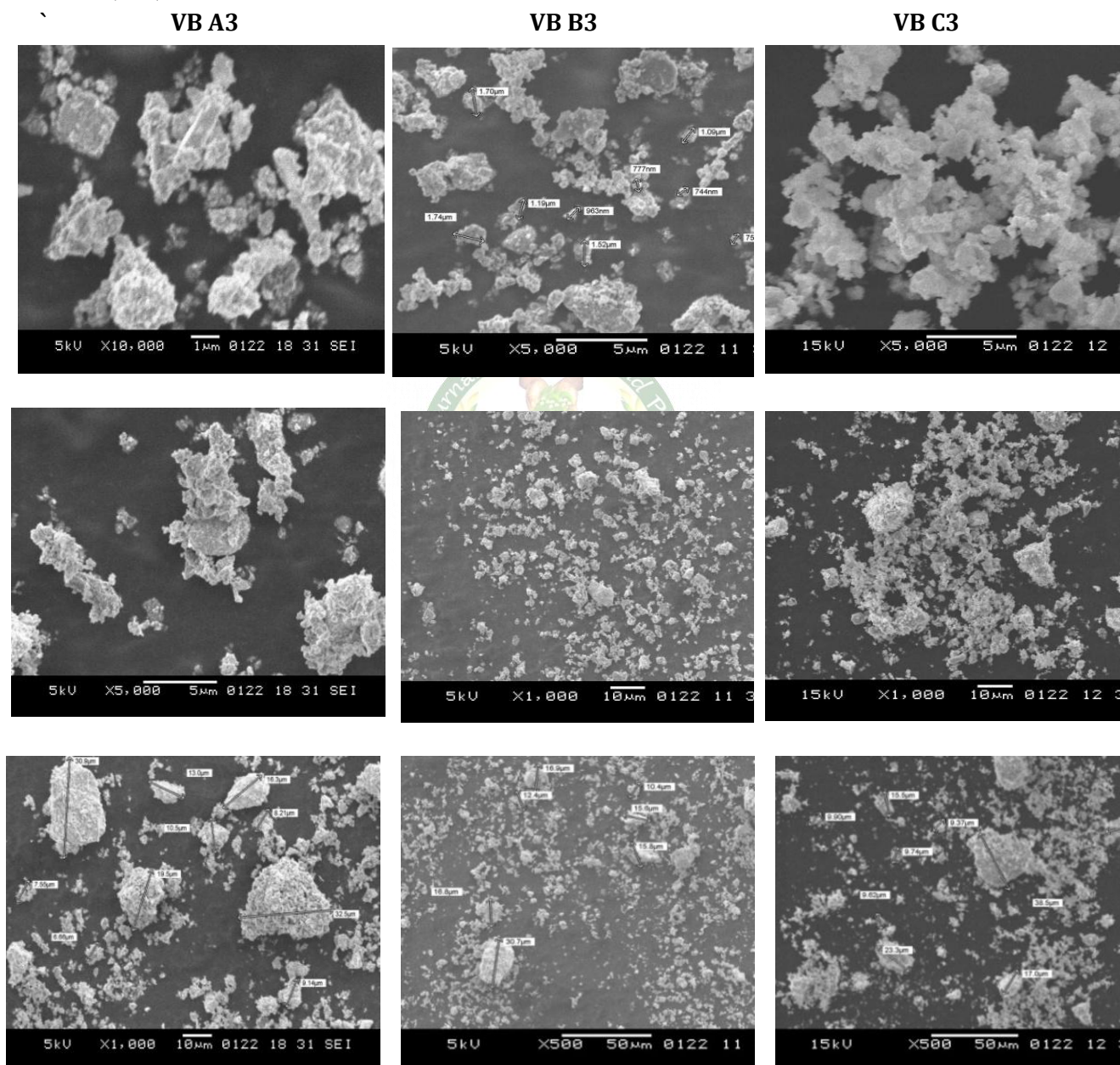
- 2<sup>nd</sup>, 4<sup>th</sup>, 7<sup>th</sup> peak with relative intensity of 100%, 76.79%, 55.26% were considered as significant at, 26.5840°, 33.8766°, 51.7761° having 3.35, 2.64, 1.76 d space value respectively.

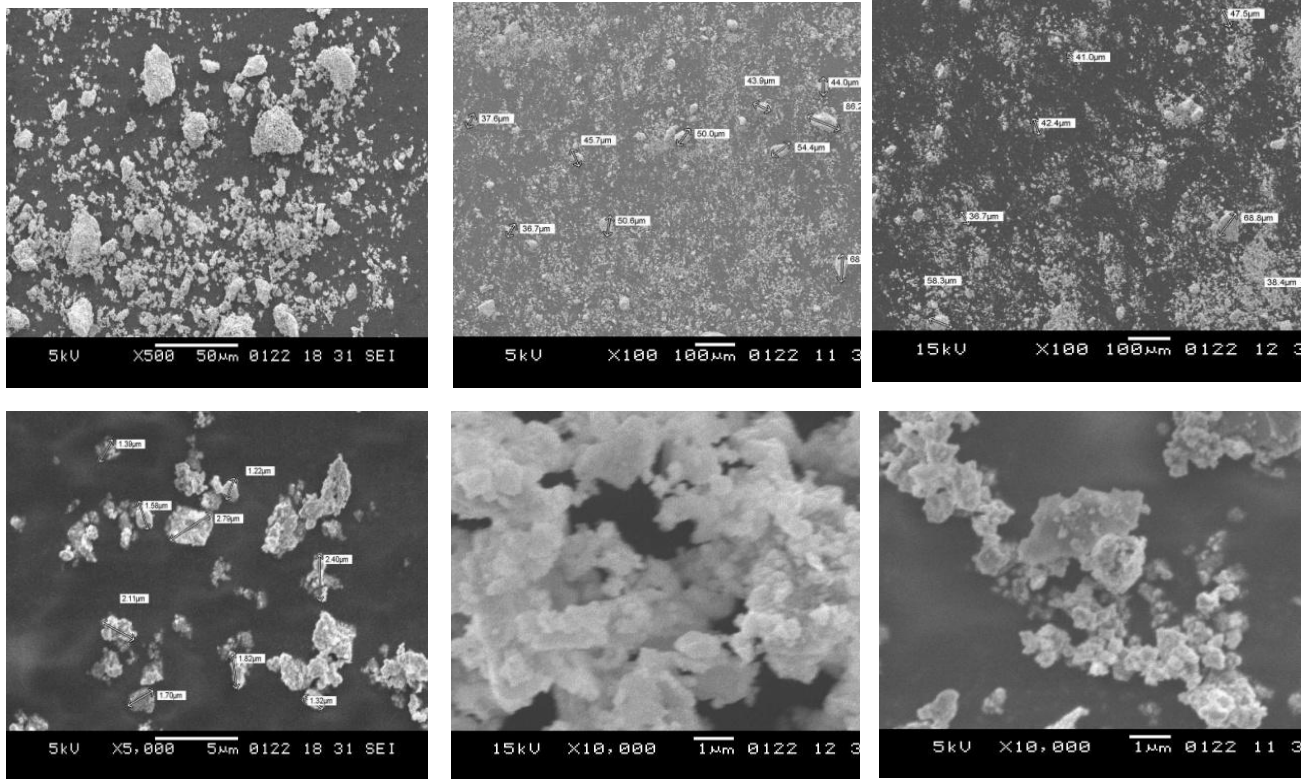
### Batch A3, B3 & C3

- Vanga Bhasma* (Batch A3, B3 & C3) peaks are compared with standard 3 theta values with ref. No.98-009-0609 confirmed the presence of Tin Oxide (Cassiterite SnO<sub>2</sub>) with hydroxide in tetragonal structure.
- Also peaks compared with standard 2 theta values with ref.No.98-009-7513 confirmed presence of Tin Sulphide (Sn<sub>2</sub>S<sub>3</sub>) with orthorhombic structure.
- Also peaks compared with standard 2 theta values with ref.No.98-063-9165 confirmed presence of Mercury Sulphide (HgS) with hexagonal structure.

### Scanning Electron Microscopy<sup>[31]</sup>

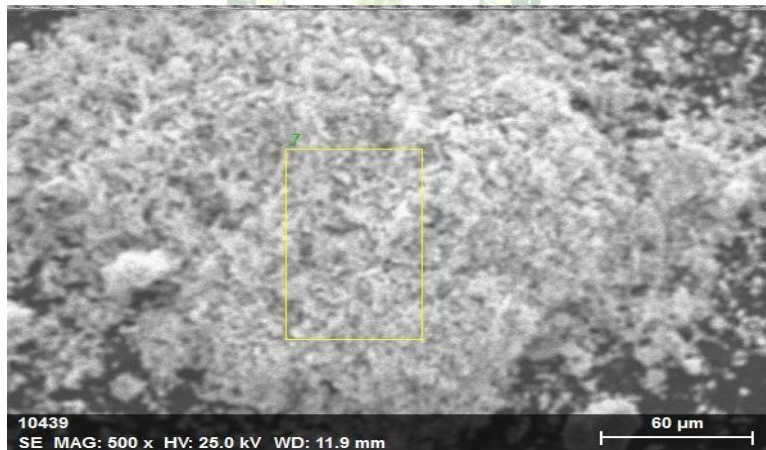
#### SEM Batch A3, B3, C3



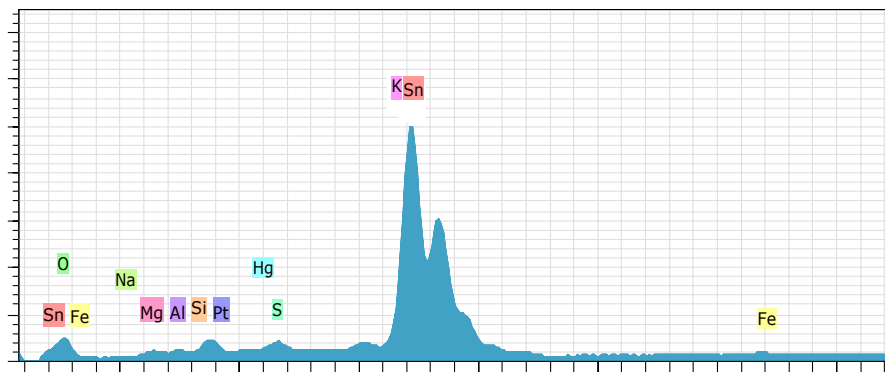


Sample A3 has a particle size of 1.22µm- 2.79µm. Sample B3 has a particle size of 744nm-953nm while sample C3 has a particle size ranging from 9.37 µm-38.5 µm.

**Energy Dispersive X-Ray Analysis<sup>[32]</sup>**  
**EDS Batch A3**



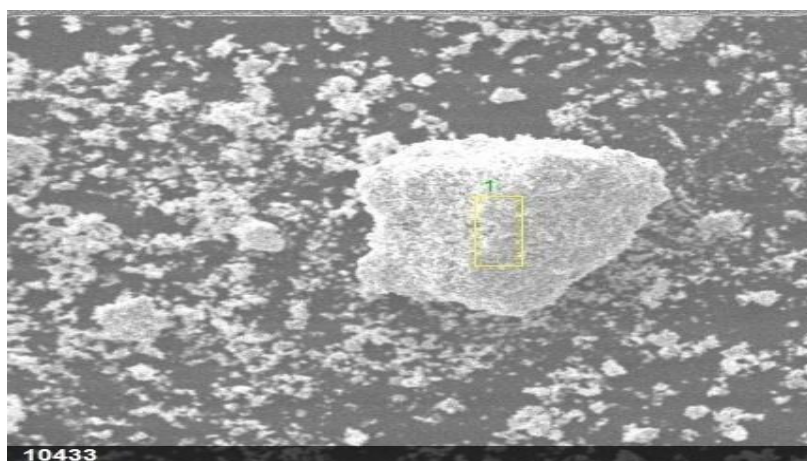
10439Date:3/4/2020 12:56:24 PM Image size: 1000 x 750 Mag:500xHV:25.0kV



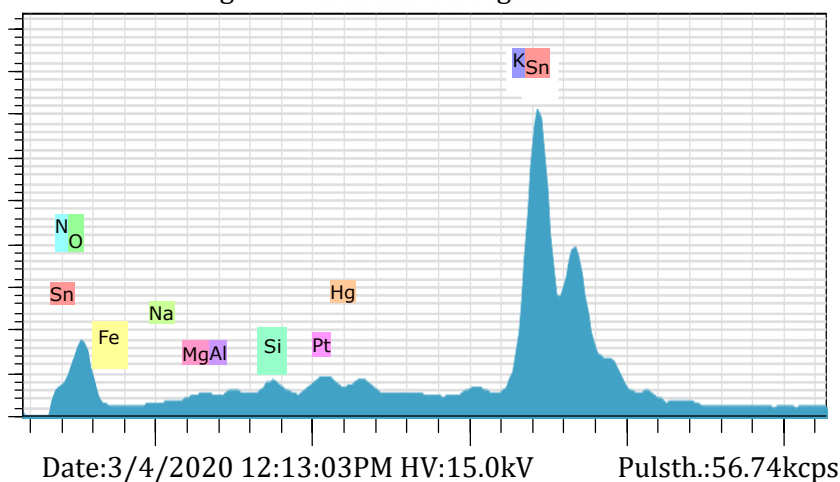
Date:3/4/2020 12:56:51 PMHV:25.0kV Puls th.:205kcps

El AN Series	unn. C norm. C Atom.	C Error (1 Sigma)	[wt.%]	[wt.%]	[at.%]	[wt.%]
Sn 50 L-series	50.95	73.34	32.30	1.52		
O 8 K-series	13.47	19.39	63.34	1.61		
Pt 78 L-series	1.87	2.69	0.72	0.08		
Hg 80 L-series	1.64	2.25	0.61	0.07		
K 19 K-series	0.80	1.15	1.54	0.05		
Fe 26 K-series	0.38	0.55	0.52	0.04		
Si 14 K-series	0.35	0.51	0.94	0.04		
S 16 K-series	0.01	0.12	0.03	0.03		
Al 13 K-series	0.00	0.00	0.00	0.00		
Mg 12 K-series	0.00	0.00	0.00	0.00		
Na 11 K-series	0.00	0.00	0.00	0.00		
Total:				69.46	100.00	100.00

**EDS Batch B3**



10433Date:3/4/2020 12:12:42 PM  
Image size:1000 x 750 Mag:500xHV:15.0kV



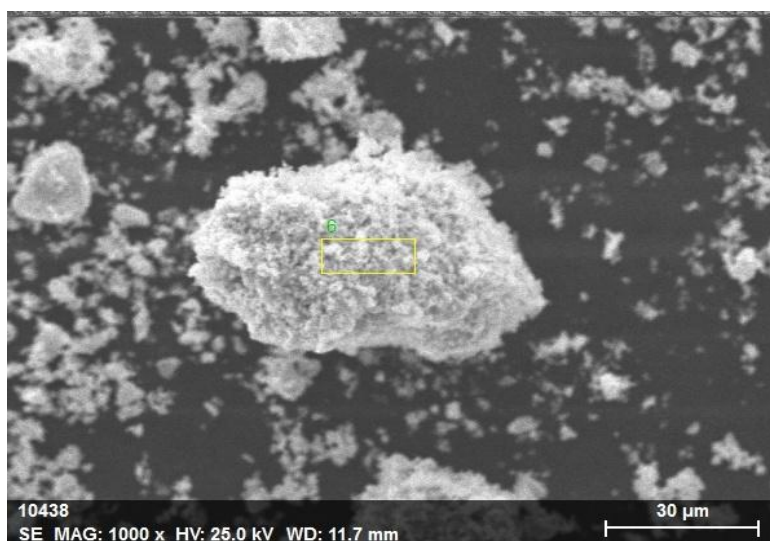


El AN Series un. C norm. C Atom. Error(1Sigma) wt. %[wt. %][at. %] [wt. %]

Sn 50 L-series	61.10	70.60	27.72	1.85
O 8 K-series	17.89	20.67	60.23	2.18
K 19 K-series	2.67	3.09	3.35	0.11
N 7 K-series	1.64	0.75	6.31	0.32
Pt 78 M-series	1.18	1.37	0.33	0.07
Fe 26 K-series	1.06	1.22	1.02	0.06
Hg 80 M-series	0.72	1.85	0.19	0.05
Si 14 K-series	0.27	0.31	0.52	0.04
S 16 K-series	0.16	0.14	0.33	0.03
Al 13 K-series	0.00	0.00	0.00	0.00
Mg 12 K-series	0.00	0.00	0.00	0.00
Na 11 K-series	0.00	0.00	0.00	0.00

Total: 86.69 100.00 100.00

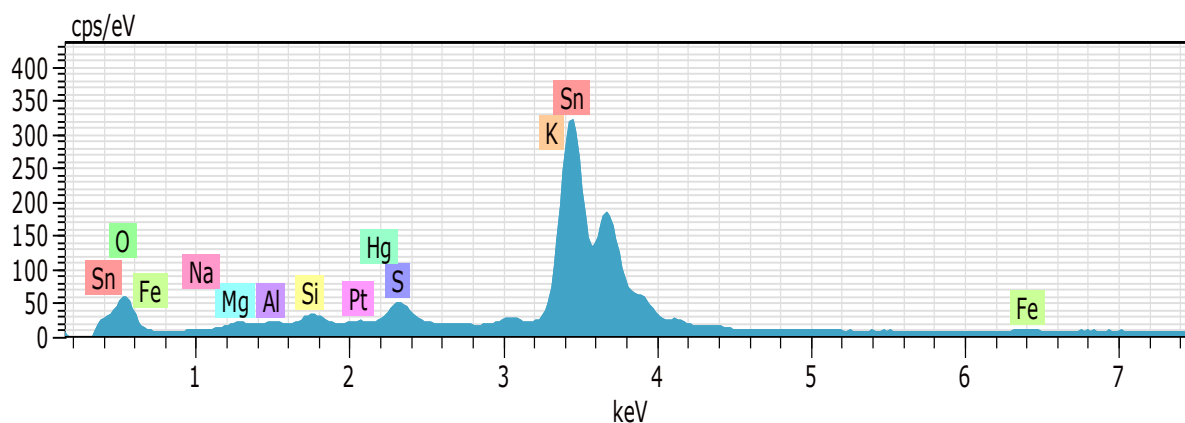
**EDS Batch C3**



10438Date:3/4/2020 12:54:14 PM Image size:1000 x 750

Mag:1000x

HV:25.0kV



Date:3/4/2020 12:54:33 PM HV:25.0kV Puls th.:266kcps

El AN Seriesun. C norm. C Atom. C Error (1 Sigma)  
[wt.%) [wt.%) [at.%) [wt.%)

Sn 50 L-series	61.10	72.66	30.72	1.85
O 8 K-series	17.89	19.46	63.54	2.18
K 19 K-series	2.67	2.94	3.35	0.11
Pt 78 M-series	1.18	1.17	0.33	0.07
Fe 26 K-series	1.06	1.02	1.02	0.06
Hg 80 M-series	0.72	2.04	0.19	0.05
Si 14 K-series	0.27	0.60	0.52	0.04
S 16 K-series	0.16	0.11	0.33	0.03
Al 13 K-series	0.00	0.00	0.00	0.00
Mg 12 K-series	0.00	0.00	0.00	0.00

Total: 86.69 100.00 100.00

EDS studies confirm the presence of 'Sn' as a major ingredient. 'O' was found as a second major ingredient in all the samples. However presence of 'O' was remarkably higher in VB. Since *Parada* was an ingredient of VB, its presence was confirmed with all the samples. Fe, Pt, K, S and Si were found in all the *Bhasma* samples;

### Antimicrobial Activity<sup>[33]</sup>

#### 1. Preparation of test Solutions/Stock solution:

The Suspensions of *Vanga Bhasma* samples A, B & C were prepared with the help of following method:

*Vanga Bhasma* sample: 100mg

Tween 80: 1g Distilled water: 10ml

So, the final concentration of the test solution obtained was- 10 mg/ml.

#### 2. Standards used in study

**Positive Control:** Cefodoxime 10mcg (Himedia Labs, Mumbai, India) was used as standard or positive control for bacteria while Flucanazol 25mcg (Himedia Labs, Mumbai, India) was used as standard or positive control for fungi in this study.

**Negative Control:** Distilled water + Tween 80

#### 3. Microorganisms:

a. Staphylococcus aureus<sup>[34]</sup>

b. Bacillus subtilis<sup>[35,36]</sup>

c. Klebsiella pneumonia<sup>[37]</sup>

d. E.coli<sup>[38]</sup>

e. Candida albicans<sup>[39]</sup>

#### Determination of Minimum inhibitory concentration Microdilution assay<sup>[40]</sup>

The minimum inhibitory concentration was defined as the lowest concentration of the compound to inhibit the growth of microorganisms (Kumar, G.S. *et al.*, 2007)<sup>[41]</sup>. The minimum inhibitory concentration values were determined by broth dilution assay of micro dilution assay. Varying concentrations of the solutions of *Bhasma* (10mg/ml, 50mg/ml, 100mg/ml) were prepared. 0.1ml of standardized test organism of Controls was equally set up by using solvents and test organisms without extract. The tube with least concentration of extract without growth after incubation was taken and recorded as the minimum inhibitory concentration.

**Table 10: Zone of Inhibition of *Vanga Bhasma* against organisms**

Sr.No	Name of Organism	Zone of Inhibition in mm								
		Sample A3			Sample B3			Sample C3		
		10Mg /ml	50 Mg /ml	100 Mg /ml	10 Mg /ml	50 Mg /ml	100 Mg /ml	10 Mg /ml	50 Mg /ml	100 Mg /ml
1	Staphylococcus aureus	11	15	18	10	14	17	10	12	17
2	Bacillus subtilis	9	13	15	10	13	18	9	11	14
3	Klebsiella pneumonia	11	14	18	12	14	19	10	13	18
4	E.coli	10	12	17	12	14	17	10	13	16
5	Candida albicans	12	15	18	13	15	19	12	14	19

**Table 11: Zone of Inhibition of control drug against organisms**

Sr. No	Name of Organism	Distilled Water + Tween80	Cepfodoxime	Fluconazole
			10mcg	25mcg
1	Staphylococcus aureus	0	24	-
2	Bacillus subtilis	0	25	-
3	Klebsiella pneumonia	0	30	-
4	E.coli	0	17	-
5	Candida albicans	0	-	28

### Statistical Study

#### ZoI of VB against Staphylococcus Aureus

In a ZoI of VB against Staphylococcus Aureus, at 95% Confidence Interval (CI), there is significant difference in the means of three different sample strengths. The sample with strength of 100mg/ml, having 17.33mm, as a mean zone of inhibition is more effective.

#### ZoI of VB against Bacillus Subtilis

In a ZoI of VB against Bacillus Subtilis, the sample with strength of 100mg/ml, having 15.66mm, as a mean zone of inhibition is more effective.

#### ZoI of VB against Klebsiella Pneumoniae

The sample with strength of 100mg/ml, having 18.33mm, as a mean zone of inhibition is more effective.

#### ZoI of VB against E. Coli

The sample with strength of 100mg/ml, having 16.66mm, as a mean zone of inhibition is more effective.

#### ZoI of VB against Candida Albicans

The sample with strength of 100mg/ml, having 18.66mm, as a mean zone of inhibition is more effective.

The findings indicate that *Vanga Bhasma* prepared with the stated reference possesses Antimicrobial property against Candida Albicans, Klebsiella Pneumoniae, Staphylococcus Aureus, E.Coli and Bacillus Subtilis in their decreasing order.

As compared to the Cefpodoxime<sup>[42]</sup> (Cephalosporin) and Fluconazole<sup>[43]</sup>, the VB preparations were found having less antimicrobial activity against all the pathogens. However, it is worth noting that this VB preparation showed antifungal activity also.

### DISCUSSION

The finding of the antimicrobial studies confirmed the action of VB (Ref. Rasatarangini 18/25-28) as an antimicrobial drug and is useful in inhibiting the growth of Candida Albicans, Klebsiella Pneumoniae, Staphylococcus Aureus, E.Coli and

Bacillus Subtilis in their decreasing order with a concentration of 100mg/ml.

As compared to the Cefpodoxime<sup>29</sup> (Cephalosporin) and Fluconazole 30, the VB preparations were found having less antimicrobial activity against all the pathogens. However, it is worth noting that this preparation showed antifungal activity also.

This outcome further supports the *Krumighna* and *Jantughna* properties (Anti-microbial activity) of *Vanga Bhasma* (Ref. Rasatarangini 18/25-28).

### CONCLUSION

The adopted methods for preparation of *Vanga Bhasma*, (Ref. Rasatarangini 18/25-28) was able to produce a *Bhasma* compatible to organoleptic parameters mentioned in the ancient texts. Formation of the small sized particles as small as a nano-particle was confirmed by SEM study.

The colour variation could be due to heat offered during the processes as well as the quality of the raw material.

1. XRD study confirms that Tin oxide is the major compound found in all the *Vanga Bhasma* samples.
2. VB (Ref. Rasatarangini 18/25-28) is an antimicrobial drug and is useful in inhibiting the growth of Candida Albicans, Klebsiella Pneumoniae, Staphylococcus Aureus, E.Coli and Bacillus Subtilis in their decreasing order with a concentration of 100mg/ml. This outcome further supports the *Krumighna* and *Jantughna* properties (Anti-microbial activity) of *Vanga Bhasma*.

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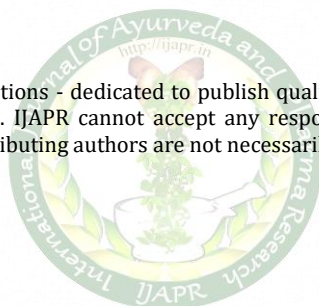
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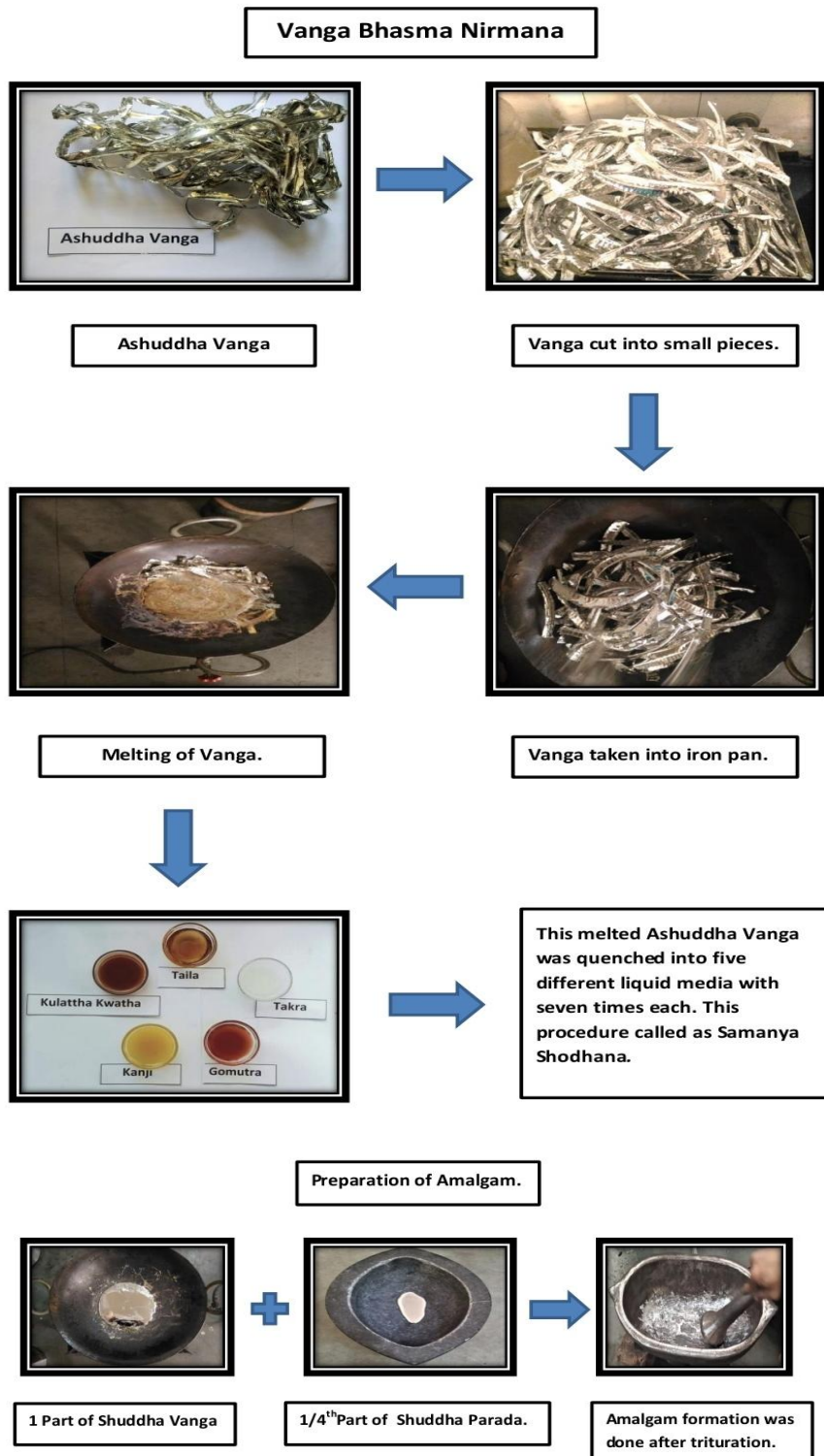
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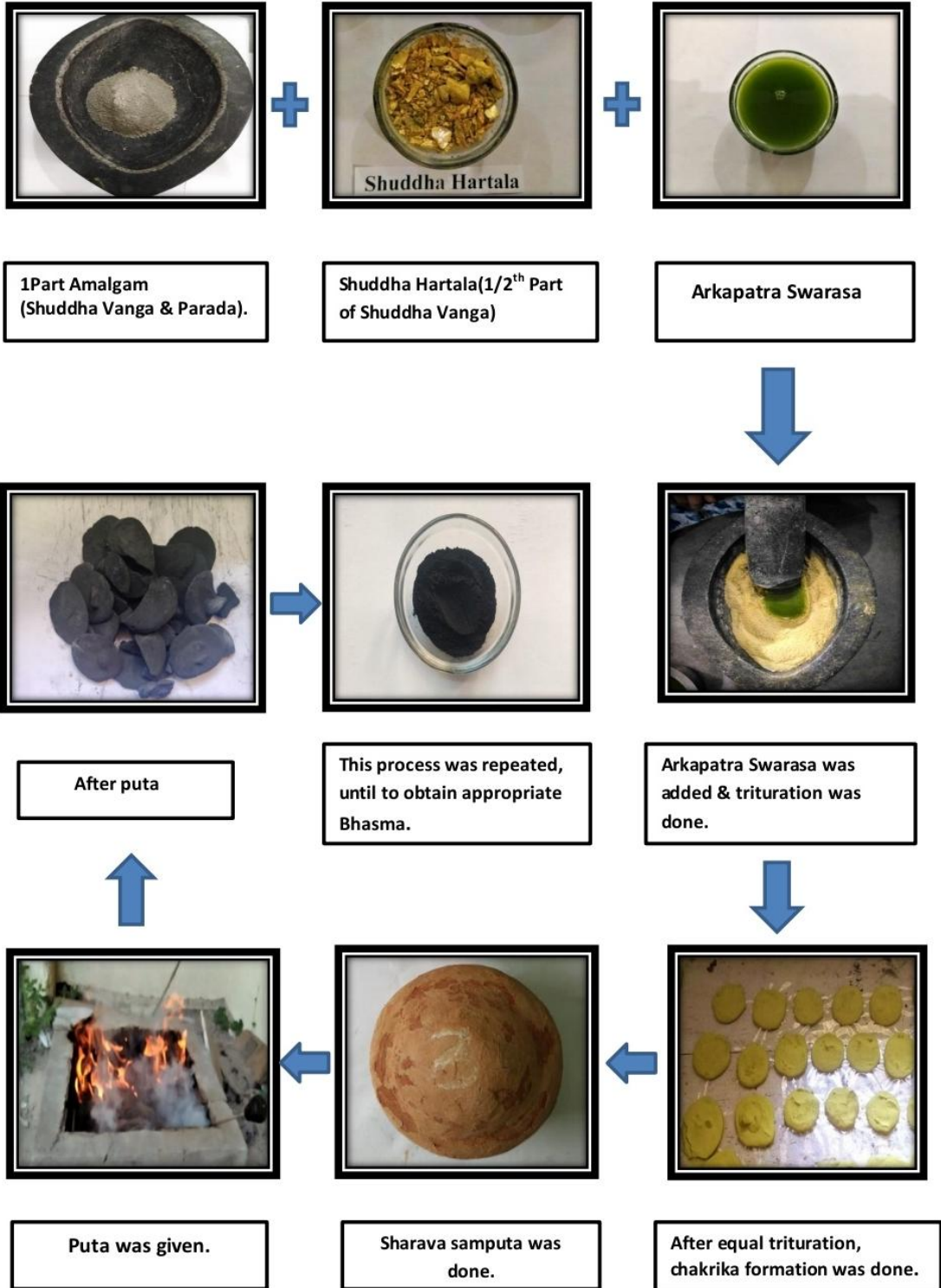
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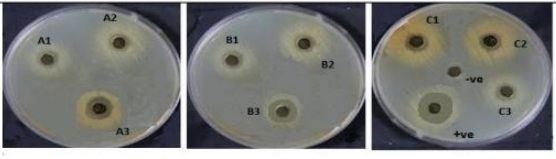

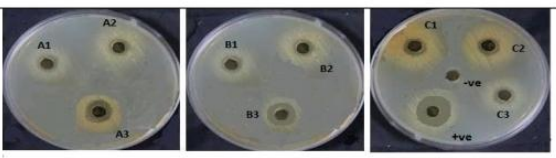
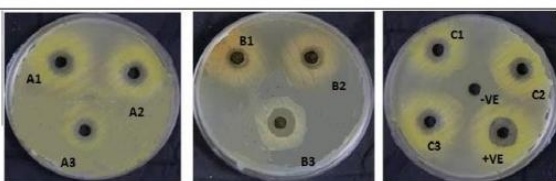




**Marana of Vanga (VangaBhasma No.3)**



**Anti- microbial Study**

Sr. no.	Micro-organism	Zone of inhibition of Vanga bhasma
1	Staphylococcus aureus	
2	E. coli	
3	Bacillus subtilis	
4	Klebsiella pneumonia	
5	Candida albicans	