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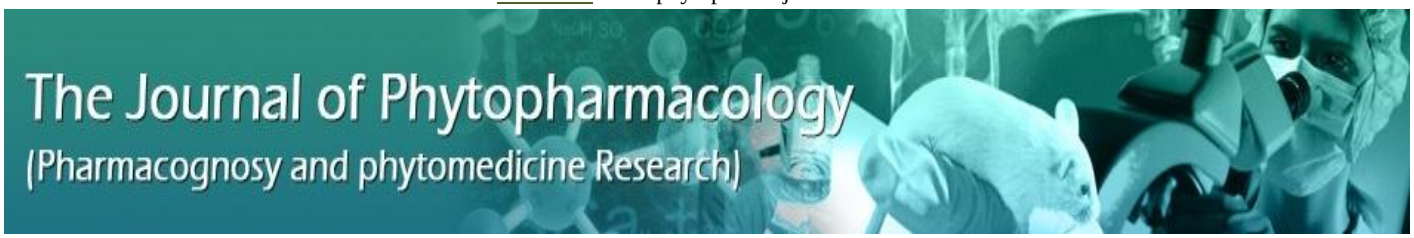

Research Article

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Standardization of *Naga Bhasma* prepared by two different *Bhavana Dravya*

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ABSTRACT

Naga Bhasma is one of the herbo-metallic preparations used in Ayurveda and claimed to possess broad spectrum of therapeutic efficacy. The Standard manufacturing Procedures of *Naga Bhasma* prepared by two methods by changing *Bhavana dravya* were developed with latex (NBAD) and leaf juice of *Calatropis procera* (NBAS). *Shuddha Manahshila* (Realgar) was taken as a *Maraka Dravya*. *Naga Bhasma* was prepared by using classical guidelines described in Rasa Ratna Samuchhaya. Total 10 Puta are required for preparation of *Naga Bhasma*. Total three batches of each group were prepared. An Average of 8.83% increase in weight of *Manahshila* was observed after *Shodhana* and 1600 ml *Ardraka Swarasa* was required for levigation. An Average of 12.57 & 18.55% increase in weight was found in NBAD and NBAS groups' respectively. An Average of 181.33 ml of latex and 182.66 mlleaf juice of *Calatropis procera* were required for *Bhavana*. Grey coloured *Naga Bhasma* was obtained after 10 Puta in EMF. Both the Bhavana dravya i.e. latex and leaf juice were found similar in view of pharmaceutical and analytical aspect, hence method of preparation of *Naga Bhasma* with *Arkapatra Swarasa* may be preferred in view of large scale manufacture, cost, labour and availability of drugs.

Keywords: *Shodhana, Marana, Bhasma, Marak Dravya, Bhavna.*

INTRODUCTION

Bhasma are organo-metallic compounds in which the metal is calcinated along with various juices,decoction of herbal ingredients to form palatable complexes[1, 2] because of usefulness in different diseased conditions since centuries without developing any major adverse effects. *Bhasma* are prepared in a well explained, generalized classical format, *Shodhana*, *Jarana* and *Marana* (programmed heating through *Puta* system). The process of *Jarana* is laboursome, uneconomic, requires isolated set up of laboratory in view of cross contamination and less safe for an operator. Hence an attempt was made in current study to develop standard operating procedure of *Naga Bhasma* without *Jarana*. *Naga Bhasma* prepared by previous researchers using arsenic trisulphide (yellow orpiment) as a *Marana Dravya*required 20 *Puta*[3]. Pravin Tate *et al*. studied the *Naga Bhasma* by adopting two different methods i.e. with *Parada, Gandhaka* and *Ashwattha, Manahshila* as *Marana Dravya*[4]. Dhirajsingh Rajput *et al*. established a standard operating procedure with the help of *Parada Pishti* and *Adhatoda vasica* Niss[5]. Four different groups of media as *Marana dravya* are mentioned in classics for *Marana*. The *Maraka Dravya* are categorised in four groups in view of superiority for *Bhasma*, which are *Rasa* (Mercurials), plant-products, sulphur & *Arilauha* sequentially in decreasing order of superiority[6]. *Manahshila* is used as *Maraka Dravya* for preparation of *Naga Bhasma* which acts as main active ingredients or as an auxiliary agent to assist the efficacy of *Naga Bhasma.* It reacts with remnant, un-reacted lead oxide (PbO) transforming it to lead sulphide (PbS) which is the least toxic form of lead[7]. The objectives of this study are to determine yield of the medicament (*Bhasma),* analyze the logical findings of *Shodhana* & *Marana,* determine physico-chemical properties of respective stage of pharmaceutical processing and evaluation of physical & chemical test parameters according to classics as well as contemporary science, to establish quality parameters, testing protocol and establish its affectivity in identifying influence of controllable factors and discuss the optimum SMP of *Naga Bhasma* preparation by two different *Bhavana Dravya* i.e. latex[8] & leaf juice of *Calatropis procera. Naga Bhasma* (incinerated Lead) is one of the well known formulation indicated in the treatment of various systemic diseases specially *Prameha* (diabetes) and is familiar as *Pramehakarikeshari*[9] i.e. one of the best drug for diabetes. In R.R.S. only latex *of Calatropis procera* has been mentioned as *Bhavana dravya* for preparation of *Naga Bhasma* but it is not possible to prepare *Naga Bhasma* with latex on large scale. Leaves of *Arka* are available in plenty. So to overcome this problem the present study has been carried out for development of Standard Manufacturing Procedure of *Naga Bhasma* with two different *Bhavana Dravya* i.e. latex & leaf juice of *Calatropis procera* and *Manahshila* (Realgar) as a *Maraka Dravya* prepared without process of *Jarana.*

MATERIAL AND METHODS

AshuddhaNaga (3kg) and *Manahshila* (6kg) with characters of acceptable quality [10, 11] as per *Ayurvedic* classics were procured from pharmacy, Gujarat *Ayurved* University, Jam*nagar*. Sesame oil, *Kulattha* (horse gram) were procured from local market and cow urine was collected from *Goshala* (cow shed), Jam*nagar*. *Latex* & leaves of *Calatropis procera* were collected from periphery of Jam*nagar*.

Preparation of *Naga Bhasma*:

Kanji (sour gruel), *Takra* (clarified butter milk) and *Kulattha Kwatha* were prepared as per classical reference from *Parada Vijnaniya*[12], *Sushurata Sutra*[13] and *Sharangadhara Samhita*[14] respectively. 99.80% chemically pure *Naga* was taken and elemental analysis of *Naga* was done by previous scholar and same batch of *Naga* was used for *Shodhana* purpose depicted in table 1[15].

Table 1: Elemental analysis of *Naga* before *Shodhana*

Element	Sn	Sb	Bi	Cu	As	Ag	Zn	Cd	Ni
Value	0.131	0.432	0.130	0.033	0.0023	0.0036	0.0004	0.0007	0.0001
Element	Ca	Al	Au	Fe	Na	P	S	Pb	
Value	0.0003	0.0001	0.0002	0.0001	0.0002	0.0068	0.0015	99.80	

Samanya Shodhana of *Naga* was done by heating it to melting and quenching in sesame oil, *Takra* (clarified butter milk), *Kanji* (sour gruel), Cow urine and *Kulattha Kwatha* (decoction of horse gram) respectively[16]. The process was repeated for three times replacing fresh liquid media every time. For *Vishesha shodhana* of *Naga*, same process was adopted using *Churnodaka* (lime water) [17]. Details of ingredients and quantity taken for *Shodhana* are depicted in table 2.

Table 2: Ingredients and their quantity

S. No	Ingredients	Latin / English name	Qty/batch	Total quantity
1	*AshuddhaNaga* (kg)	Lead	1	3
2	*Tila Taila* (*l*)	*Sesamum indicum* Linn.	3	9
3	*Takra* (*l*)	Butter milk	3	9
4	*Kanji* (*l*)	Sour gruel	3	9
5	*Go Mutra* (*l*)	Cows urine	3	9
6	*Kulattha Kwatha* (*l*)	*Dolichos biflorus* Linn.	3	9
7	*Churnodaka(l)*	*Lime Water*	7	49

Average of observed test parameters during *Samanya* and *Vishesha Shodhana*of *Naga* in different media is mentioned in table-3.

Table 3: Average data observed during *Samanya* and *Vishesha Shodhana* of *Naga*

S. No	Media	Avg. time (min)	Avg. Wt. (g)		Loss of wt (Avg. in g)	% of loss (Avg.)
			BS	AS		
1	*Tila*	4.49	1000	989.65	10.35	1.035
2	*Takra*	5.20	989.65	977.35	12.30	1.24
3	*Kanji*	5.59	977.35	967.53	13.83	1.41
4	*Gomutra*	6.12	963.52	948.33	15.19	1.57
5	*Kulatha*	6.89	940.33	924.29	16.04	1.70
6	*Churnodaka*	7.12	926.29	909.11	17.18	1.85

BS-Before *Sodhana*, AS- After *Sodhana*

Shodhana of *Manahshila* was performed as per reference of *Rasatarangini* (table -4) [18].

Table 4: Observation of *Manahshila Shodhana*

Batch	Qty. of *Ashodhit Manhshila* (kg)	Total Number of *Bhavana*	Qty. of (*Ardraka Swarasa*) in ml	Qty. of *Shodhita Manahshila* (kg)
1st	2	7	1600	2.14
2nd	2	7	1600	2.15
3rd	2	7	1600	2.18

Naga Patra (Lead sheets) were prepared from *Shodhita Naga* by rolling press machine and cutting into 4 X 1 inch (length X breadth). Then paste of *Shuddha Manahshila and latex of Calatropis procera* was prepared by levigation method and smeared over both sides of *Naga Patra* and dried in sun light. Next day dried *Naga Patra* was weighed, collected in *Sharava* (an earthen saucer, covered by another earthen saucer and the junction was sealed by mud smeared cotton cloth) and was allowed for complete drying. Next day *Sharava* were subjected for *Puta* in Electric Muffle furnace till attainment of 500°C, followed by self cooling inside furnace. Next day after self-cooling (*Swangsheet*) *sharava* were collected, *Chakrika* (incinerated pellets) were removed, weighed and ground to fine powder.Again1/20th part *Manahshila* as that of weight of *Naga* after 1st*Puta*was added to this powder and later on levigated with *latex of Calatropis procera* followed by preparation of pellets and incineration. Same procedure was repeated 10 times for each batch (table-5). The same procedure was followed for NBAS sample using leaf juice of *Calatropis procera*.

RESULTS

The study inferred that an average of 7.57% and 1.7% loss was observed after *Samanya* and *Vishesha Shodhana* respectively. An average of 1600 of *Ardraka Swarasa* (ginger juice) was required for levigation of 2 kg of *Manahshila* for duration of trituration of 7.35 hrs in stone end runner for 7 times. An average of 8.83% increase in weight of *Manahshila* was observed after *Shodhana*. *Marana* of *Naga* by latex of *Calatropis procera* was comparatively early and the quality of *Bhasma* was also found to be comparatively better as that of *Bhasma* prepared by leaf juice of *Calatropis procera.* Average of 12.57 & 18.55% increase in weight was found in NBAD and NBAS groups respectively. An Average of 181.33 ml of latex and 182.66 ml leaf juice of *Calatropis procera* were required for *Bhavana*. Grey coloured *Naga Bhasma* was obtained after 10 *Puta* in EMF. In the physico-chemical analysis, *Shuddha Manahshila* has slightly more ash value than that of *Ashuddha* (6.12% and 7.25% respectively. Comparatively increase in loss on drying in *Shodhita Manahshila* as that of *Ashuddha Manahshila* (0.22% and 0.13% respectively) was observed. Average 0.40, 99.91 6.33, 8.46 and 0.19, 99.9, 16.58, 6.30 LOD, ash value, acid insoluble ash, pH was observed in NBAD and NBAS group respectively.

Table 5: Details of *Marana* of *Naga*

S. No.	Ingredients	NBAD			NBAS		
1st*Puta*		Batch 1	Batch 2	Batch 3	Batch 1	Batch 2	Batch 3
	Naga(g)	300	300	300	500	500	500
	ShuddhaManahshila (g)	300	300	300	500	500	500
	Latex / Leaf Juice (ml)	200	190	200	250	230	250
	Wt.of dried *NagaPatra* before *Puta*	629	620	639	1019.4	1012	1008.2
2nd*Puta*	*Naga*(g) after 1st*Puta*	390.6	399.28	403.25	642.3	652.8	634.6
	ShuddhaManahshila (g)	19.53	19.96	20.16	32.11	32.64	31.73
	Latex / Leaf Juice (ml)	160	170	170	180	180	170
	Wt.of *Chakrika* before *Puta*	424.55	429.25	434.50	697.7	703	692
3rd*Puta*	*Naga*(g) after 2nd*Puta*	400.25	404.58	409.78	640.7	643	638.1
	ShuddhaManahshila (g)	20.01	20.22	20.48	32.03	32.15	31.90
	Latex / Leaf Juice (ml)	170	180	190	160	170	160
	Wt.of *Chakrika* before *Puta*	435.25	433.98	439.26	681	687.4	688
4th*Puta*	*Naga*(g) after 3rd*Puta*	401.20	399.25	397.21	634.9	637.5	640
	Shuddha Manahshila (g)	20.06	19.96	19.86	31.74	31.87	32.0
	Latex / Leaf Juice (ml)	180	180	190	160	170	160
	Wt.of *Chakrika* before *Puta*	429.30	430.10	426.59	677	668.8	679
5th*Puta*	*Naga*(g) after 4th*Puta*	394.30	397.2	393	630	628.4	636.9
	Shuddha Manahshila (g)	19.71	19.86	19.65	31.5	31.42	31.84
	Latex / Leaf Juice (ml)	180	170	190	170	180	190
	Wt.of *Chakrika* before *Puta*	420.8	423.4	424	675	673.15	671.2
6th*Puta*	*Naga*(g) after 5th*Puta*	385.2	383	389	625	624.12	620
	Shuddha Manahshila (g)	19.26	19.15	19.45	31.25	31.20	31
	Latex / Leaf Juice (ml)	180	170	190	180	180	190
	Wt.of *Chakrika* before *Puta*	410.15	416.6	411	672.3	672	669.2
7th*Puta*	*Naga*(g) after 6th*Puta*	380.25	379	376	620.3	620	625.10
	Shuddha Manahshila (g)	19.01	18.96	18.80	31.01	31	31.25
	Latex / Leaf Juice (ml)	160	180	190	180	180	170
	Wt.of *Chakrika* before *Puta*	410.23	412	416	662	689.8	676
8th*Puta*	*Naga*(g) after 7th*Puta*	369	370.19	368	614	608.6	631
	Shuddha Manahshila (g)	18.45	18.50	18.4	30.7	30.43	31.55
	Latex / Leaf Juice (ml)	180	170	180	170	180	170

	Wt.of *Chakrika* before *Puta*	400.2	402	405.3	656.9	662.3	665
9th*Puta*	*Naga*(g) after 8th*Puta*	351	356.9	359.2	609.1	613.5	617
	Shuddha Manahshila (g)	17.55	17.84	17.96	30.45	33.56	38
	Latex / Leaf Juice (ml)	190	180	190	180	180	190
	Wt.of *Chakrika* before *Puta*	381	387.7	385.3	642.8	649	653
10th*Puta*	*Naga*(g) after 9th*Puta*	350.5	353	359.3	595.0	606.8	610
	Shuddha Manahshila (g)	17.52	17.65	17.96	29.75	30.34	30.5
	Latex / Leaf Juice (ml)	190	180	190	180	190	180
	Wt.of *Chakrika* before *Puta*	379.1	380	384.4	640	645.5	654
	Naga(g) after 10th*Puta*	334	339.2	340	587	592	599.35

Table 6: Results of the physicochemical analysis of *Ashuddha* & *Shuddha Manahshila*

S. No.	Parameter	*AshuddhaManahshila*	*ShuddhaManahshila*
1.	Loss on drying at 110ºC	0.13	0.22
2.	Ash value	6.12	7.25
3.	Acid insoluble ash	0.79	0.41
4.	Water Soluble Ash	0.04	0.04
5.	Loss on ignition at 450ºC	91.19	92.12
6.	pH	8.11	7.32

Table 7: Organoleptic characters of *Naga Bhasma*

S. No.	Parameters	NBAD	NBAS
1.	*Shabda*	No metallic sound	No metallic sound
2.	*Sparsha*	No course particle	No course particle
3.	*Rupa*	Grey colour	Grey colour
4.	*Rasa*	Tasteless	Tasteless
5.	Gandha	Not specific	Not specific

Table 8: Results of the physicochemical analysis of *Naga Bhasma*

S. No.	Parameter	*Naga Bhasma* (NBAD)			*Naga Bhasma* (NBAS)		
		I	II	III	I	II	III
1.	Loss on drying at 105ºC	00.48	00.39	00.32	00.24	00.19	00.14
2.	Ash value	99.95	99.90	99.90	99.90	99.95	99.85
3.	Acid insoluble ash	06.4	05.98	05.62	16.58	16.71	16.45
4.	pH	8.29	8.22	8.87	6.12	6.46	6.34

DISCUSSION

Naga is one among *Rasavarga dravya* used in *Ayurvedic* formulations which is mentioned under heavy metals[19] but used strictlyin therapeutics only after classical processing like *Shodhana, Jarana and Marana*. If *Naga* is used without proper *Shodhana* and *Marana*, it causes various diseases (*Naga-Dosha*) like *Kushtha, Gulma, Ruja, Meha, Pandu, Jwara, Tridoshaprakopa Roga*[20], *Mrityu*[21] *Udara*[22] *Kilasa, Sandhishula*[23], *Pakshavadha, Anaha, Amsashotha, Avabahuka, Shula*; *Kandu, Anilasada*[24] etc. All *Acharya* advised to take *Swarna Bhasma* (incinerated gold), *Haritaki (Terminalia chebula Lin.),* and *Sita* (sugar) internally for 3 days to get relief from untoward effects of improperly processed *Naga*[25-27]. Author of *Rasatarangini* mentioned *Vishatinduka* (*Strychnos nuxvomica*) as an antidote for *Nagadosha*[28]. In *Rasahridayatantra,*cow urine, *kutki* (*Picrorrhiza kurroa*) and root of *Karavellaka* (*Momordia charantia*) are advised as a remedy for untoward effect of *Nagadi Kalankita Rasa* (a formulation of Lead) [29]. *Shodhana* is the process in which *Mala* (physical and chemical impurities) are removed by means of *Peshana* (trituration) etc[30]. The toxic substances are minimized or removed and the desired qualities are imparted to the material. In *Ayurveda*, a great emphasis is placed on *Shodhana* (purification) and detoxification of metals and other minerals. *Shodhana* is first and most important step before preparing their *Bhasma* which is of 2 types i.e. *Samanya Shodhana* (common method) and *Vishesha Shodhana* (specific method of *shodhana*). *Samanya Shodhana* of *Naga* is the general method of *Shodhana* for the *Loha Varga*. *Samanya Shodhana* involves heating the metal to red hot stage or up to complete melting followed by quenching for either three or seven times in each liquid media which are sesame oil, clarified butter, cow urine, sour gruel and decoction of horse gram respectively. *Vishesha Shodhana* involves similar procedure conducted with specific liquid media for different metals. Conversion of *Naga*in to *Bhasma* form is quite difficult process due to its low melting point.

As per AFI for *Samanya Shodhana* of *Naga*, process of *Dhalana* is mentioned to repeat thrice[31]. In present study 99.8 % chemically pure 3 kg *Naga* (lead) was taken for *Shodhana* process and minimum 1 litre liquid media was found sufficient to completely immerse material during quenching. During quenching in sesame oil, *Naga* caught fire

which was continued even till molten stage. Pungent smell, hissing sound and black fumes were observed after quenching and iron ladle turned yellow in colour. Flame was noticed after quenching in *Takra* as well. Hence flame, black fumes after 1st quenching in Sesame oil and *Takra* are likely due to burning of residual oil and Ghee respectively. Some part of *Naga* got converted into yellowish powder after each quenching. Melting time of *Naga* was gradually increased after *Shodhana* in each media. Average time taken for complete melting (apart from powdered part)was 4.49, 5.20, 5.59, 6.12, 6.89 and 7.12 min. for *sesame oil*, clarified butter milk, sour gruel, cow *urine*, decoction of horse gram and lime water respectively. Shining of *Naga* was decreased after quenching in *Kanji* and *Gomutra*, while it was significantly decreased after *Shodhana* in *Kulatthakwatha*. *Naga* acquired silver luster after each quenching. There was persistently increase in percentage weight loss after quenching in each media i.e. an Average1.03, 1.24, 1.41,1.57,1.70,1.85 % weight loss after *Shodhana* in sesameoil, clarified butter milk, sour gruel, cow urine, decoction of horse gram and lime water respectively. An average of 7.57% and 1.7% loss was observed after *samanya* and *Vishesha Shodhana* respectively. Gradual increase in percentage loss after *shodhana* in each media is due to conversion of *Naga* in to oxide (powder) form and loss obtained during filtration. Previous study reported that impurities of *Naga* may get attracted towards the chemical components present in the media forming a bond with them and they may get separated hence each time fresh liquid media was taken[32].Gradual increase in melting time was observed while quenching which is due to oxidation of Lead and partial conversion of *Naga* into powder (PbO). Powdered *Naga* absorbs the liquid media during quenching which upon subsequent heating to molten state, delays rise of temperature of mixture and prolongs time taken to melt. Powder form of *Naga* settles at the bottom of the iron ladle thus changing heat transfer from iron ladle to metallic lead causing delay in achievement of melting temperature[33]. Another study reported that all the media utilized for *Shodhana* purpose form soluble-chelates with metals, leading to their removal resulted in the increase of relative mass percentage of lead from 97.13% in the raw material to 98.98%[34].

In present study *Marana* process was done using two liquid media. Classically latex *of Calatropis procera* is used for *Bhavana* during *Marana* process, but in large scale production getting sufficient quantity of latex is difficult. Hence to overcome this, present study was conducted to find out effect of both liquid media for *Marana* on pharmaceutico- analytical profile of *Naga Bhasma*.

Arsenical compounds have a long and remarkable history of pharmacological utilities and traditional practices[35]. Arsenical compounds are intentionally added to *Ayurvedic* formulations as main active ingredients or as auxiliary agent to assist the efficacy of herbal drug. Temperature during *Marana* probably facilitates conversion of arsenic sulphide (As_2S_2) to arsenic oxide (As_2O_3) and thus conversion of PbO to PbS. The boiling point of arsenic oxide (As_2O_3) is about 465°C and hence would have vaporised during the incineration cycle where PbS is known to react with PbO when heated, leading to formation of metallic lead and sulphur dioxide. An addition of realgar during each incineration cycle may probably be aimed at suppressing the above reaction. The addition of realgar in excess quantity reacts with any unreacted PbO transforming the same to PbS. This ensures that the intermediates do not contain elemental lead. These finding denotes the importance of *Manahshila* in the preparation of *Naga Bhasma* which assist the formation of PbS and ensure the safety aspect of *Naga Bhasma* because intermediate compound PbO is more toxic as compared to lead sulphide[36]. In *Shodhana* of *Manahshila*, levigation with *Ardraka Swarasa* changed the orange colour of *Manahshila* powder to reddish yellow. In the physico-chemical analysis, *Shuddha Manahshila* has slightly more ash value than that of *Ashuddha* (6.12% and 7.25% respectively, Table-6). Observed increase may be due to addition of inorganic components of *Ardraka Swarasa*. Comparatively increase in loss on drying in *Shodhita Manahshila*as that of *Ashuddha Manahshila* (0.22% and 0.13% respectively) was found which is probably due to hygroscopic nature of total solid of *Ardraka Swarasa*. An average of 1600 of *Ardraka Swarasa* (ginger juice) was required for levigation of 2 kg of *Manahshila* for duration of trituration of 7.35 hrs in stone end runner for 7 times. An average of 8.83% increase in weight of *Manahshila* was observed after *Shodhana*.

Marana is a process in which *Shodhita Dravya* converted into *Bhasma* by applying the procedure of *Bhavana* and specified heating protocol. The main objective of *Marana* is to achieve *Rasibhavana* of *Loha* (*Dhatu* etc *rasavarga dravya*) i.e. to reduce the metal mineral in to finer particle so that they could get absorbed in to the system, mix with the *dhatu*like *Rasa, rakta*, etc., get assimilate; metabolise and excreted easily, make them compatible with bodily structures and thus produce their desired effects without producing harmful effects.

Bhavana is also a key procedure in *Marana* of metals. Therapeutic efficacy of *Naga Bhasma* changes with change in the method of preparation e.g. *Naga Bhasma* prepared by *Bhavana* of *Ahiphen* (*Papaver somniferum*) possess more aphrodisiac property than the *Bhasma* prepared from *Manashila* and the same when prepared using *Gandhaka* and *Vasa (Adhatodia vasaica* Niss*.)* will be more effective in skin diseases[37]. *Latex of Calatropis procera* was vigorously described for *Shodhana* and *Marana* of various metals in classics[37]. *Bhavaprakasha* described that the milky latex of *Calatropis* possess *Tikta* (bitter), *lavana* (salty) rasa, *ushnavirya* (hot in potency), *snigdha* (unctuous), *tikshna* and *laghu* (light) *guna*[38]. Because of its *tiknshnatva* it helps in reduction of particle size of metals and conversion in to nano particles[39]. Wide use of *Latex of Calatropis procera* as a *Bhavnadravya* for *Marana* of various metals in our classics may be attributed to these properties. There are more than 133 references of use of *Arka* (*Calatropis procera*) in pharmaceutical processes of *Rasavarga Dravya* (Mercurials and metallo mineral drugs) among which 55 references are for use in various processing of *Dhatu* (metals). Latex of *Calatropis procera* is most frequently used plant part of *Arka* and coincides with modern green synthesis of particles[40]. For the preparation of both samples of *Naga Bhasma* (NBAD & NBAS), *Manahshila* was used as *Maraka dravya*. In first *Puta* Lead sheet was prepared to expose maximum surface for *Marana*. In first *Puta*, sequentially *temperature* was raised in Horizontal Electric Muffle furnace till 500°C followed by period of self cooling. Maximum quantity of sheets of *Naga* got converted to powder while little fraction turned to black coloured bolus and plenty of silvery crystals of arsenic got deposited on inner side of upper *Sharava* and on the upper layer of pellets. In subsequent *Puta, only* 1/20th part of *Manahshila* was added as that of material obtained after each incineration cycle. After second *Puta*, pellets were hard, greyish black in colour, hard in touch and with absence of crating/ roughness /friction during grinding but minute shining were observed in NBAD group. In NBAS group, after second *Puta* pellets were hard, granular and black in colour with some metallic particles (*Utthapita Naga)* observed during trituration, having shining at the cut surfaces. In NBAD after 3rd *Puta*, pelletswere found soft and whitish colour was appeared on the surrounding areas whereas in NBAS samples, pellets were more hard, granular, having metallic sound and black in colour but more fragile, some metallic particles of *Utthapita Naga* and shining at the cut surface were observed. Greyish black colour, hard pellets were found till 5th*Puta*. In NBAD Greyish coloured, fragile and soft pellets were found till 6th*Puta* and which didn't shown shining. Pellets were soft in touch, easily breakable and without shining at the cut surface after 8th*Puta* in NBAD group and passed *Bhasma Pariksha*. In NBAS group, pellets were somewhat hard as that of NBAD group but after10 *Puta*, it passed *Bhasma Pariksha* like *Varitar, Apunarbhava, Niruttha*. Colour of *Bhasma* was greyish black (*Kapot Varna*). *Naga Bhasma* may be considered as sulphide form of lead (PbS) which is greyish black in colour[41], along with traces of other elements. An average of 29.66 & 30.17 % increase in weight of all three batches were found in NBAD and NBAS groups respectively.

In addition, *Shodhana* facilitates further pharmaceutical processing of *Marana*. The use of realgar during each incineration cycle enables the conversion of PbO to PbS along with elimination of arsenic as arsenic oxide vapours. Repeated *Putana* process plays predominant role in attaining monodispersity of the *Bhasma* particles. The existing *Bhasma* preparation techniques follow different procedures for *Naga Bhasma* and the protocol may differ in terms of raw materials utilised, variety of plants used and number of calcination cycles performed [3-5]. Protocol of SOP of *Naga Bhasma* prepared with changing liquid media i.e. latex and leaf juice of *Calatropis procera* provide important pharmaceutico analytical data for industrial production of *Naga Bhasma* without process of *Jarana* with comparatively more safety in pharmaceutical processing, economic method and minimization of chances of cross contamination.

Marana of *Naga* by latex of *Calatropis procera* was comparatively early and the quality of *Bhasma* was also found to be comparatively better as that of *Bhasma* prepared by leaf juice of *Calatropis procera*, however as *Bhasma* prepared with leaf juice of *Calatropis procera* (NBAS)has almost similar pharmaceutical and physico-chemical profile compare to NBAD, hence it can be postulated that both of them might have similar therapeutic value which can be proved only with pharmacological and or clinical studies. The present study has followed the standard preparation protocol mentioned in *Rasaratnasamuchaya* and *Anubhut* method and scientifically validated the steps involved in the preparation. The preparative technology of *Naga Bhasma* is complex, laborious and time consuming procedure and such a type of cost effective methods with scientific insights should be adopted in present prospective with the justification of Good Manufacturing Practices guidelines and safety aspect.

CONCLUSION

Although *Naga Bhasma* was prepared early by latex of *Calatropis procera* and the *Bhasma* was comparatively better on basis of organoleptic characteristics, the study denotes that equal number of *Puta* i.e.10 *Puta* are sufficient for preparation of *Naga Bhasma* with both liquid media (latex and leaf juice of *Calatropis procera*) without process of *jarana* and there were insignificant differences in pharmaceutical and analytical studies of *Naga Bhasma* prepared by either methods.However alternative method of *Naga Marana* with *Leaf juice of Calatropis procera* without *Jarana* may be preferred over *Naga marana* by latex of *Calatropis procera* in view of large scale industrial production, economy and labour.

Conflits of Interest

No conflicts of interest.

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