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Research Article

A COMPARATIVE PHARMACEUTICO-ANALYTICAL STUDY OF *KASEESA SHODHANA* WITH DIFFERENT METHODS

Bhanderi Radhika[1]*, Gohil Kunal M[2], Kantariya Bhumi[3]

*[1]M.Pharm (Ayu) Scholar, [2]Senior Lecturer, [3]Lecturer, Department of Rasashastra and Bhaishajya Kalpana, ITRA-Pharmacy, India.

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ABSTRACT

Kaseesa is classified under two groups - *Uparasa* and *Upadhatu*. It is a mineral drug used in therapeutics. Hence *Shodhana* should be done before further use in as therapeutics or for preparation of *Bhasma.* The objective of this article is to compare pharmaceutical and analytical aspects of *Ashuddha Kaseesa* and *Shuddha Kaseesa* (*Sodhana* with different methods). **Material and Methods:** Present study aimed to compare different *Shodhana* methods of *Kaseesa* which is by *Bhavana* in *Bhringaraja Swarasa*, *Swedana* in *Bhringaraja Swarasa*, *Bhavana* in *Nimbu Swarasa* and *Bhavana* in citric acid solution rather than *Nimbu Swarasa*. The preliminary physico-chemical analysis was performed simultaneously for the *Ashuddha* and different samples of *Shuddha Kaseesa* (*Shodhana* by different methods). The total Iron contain and total inorganic contain were carried out by EW-XRF Spectroscopy technique. **Results:** After *Shodhana* with different methods average yield of *Shuddha Kaseesa* was 99.72%, 108.33%, 96% and 84% method A, B, C and D simultaneously. Total iron concentration of AK was 26.28. Significant physical changes have been observed after *Shodhana*. Which reflects in BBK, SBK, MNK and MCK has 23.52, 21.96, 26.6 and 41.46 percentage of total iron contain. **Conclusion:** Pharmceutico-analytical study revealed that there were major physical and chemical changes occur after *Shodhana* in *Kaseesa*. Each method and liquid media led to major physical and chemical changes in *Kaseesa*. Which also reflects in total iron contain of *Shuddha Kaseesa* as compare to *Ashuddha*.

INTRODUCTION

Kaseesa (Ferrous Sulphate) is widely mentioned in Ayurvedic classics, Ayurvedic pharmaceutics and therapeutics as well as it is widely practiced clinically in the management of various disease conditions internal as well as external.[1] It is abundantly available in India as well as all over the globe. Thus, it is economic and considerably safer mineral if processed properly and used justifiably. It is a major source of iron along with sulphate mainly. In view of its origin and nature, it is secondary mineral found along with minerals and it can be artificially manufactured. Natural variety of *Kaseesa* may contain other trace elements also like manganese, calcium etc. It also has been prescribed in modern medicine as hematic drug but here it is used without any *Shodhana Samsakara*, because in modern science only chemical purity is considered as standard. Thus, *Kaseesa* is among few drugs that is used by almost all medicinal sciences in the world.

Different methods of *Shodhana* of *Kaseesa* have been mentioned in classics, which have different clinical implications hence study was planned to evaluate pharmaceutico-analytical differences among methods of *Kaseesa Shodhana* to generation of data for postulation of change after *Shodhana* and change among different methods of *Shodhana* of *Kaseesa*.

Till date, there is no comparative study has been conducted for *Kaseesa Shodhana* by various different methods. So, here in this study, comparative pharmaceutical process and analytical

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profile of *Ashuddha* and *Shuddha Kaseesa* by following mentioned methods are executed.

The study may find helpful in phamaceutico-analytical evaluation of difference in *Ashuddha Kaseesa* (AK) and *Kaseesa* after *Shodhana* and differentiation of chosen methods of *Shodhana* as well as differentiation among analytical profiles of *Shuddha Kaseesa* prepared with *Bhavana* with *Bhringaraja Swarasa* (BBK), *Swedana* in *Bhringaraja Swarasa* (SBK), *Maradana* (*Bhavana*) in *Nimbu Swarasa* for one day (MNK) and one modified method *Maradana* (*Bhavana*) in citric acid solution for one day (MCK). Thus, study will help in obtaining data for manufacturing processes of these methods and also probable analytical profile.

MATERIALS AND METHODS

The authentic material of *Kaseesa* was obtained from I.T.R.A. Pharmacy, Jamnagar. *Shodhana* process was carried out in Department of Rasashastra and Bhaishajya Kalpana I.T.R.A. Analytical work was performed in Department of Pharmaceutical Chemistry I.T.R.A.

Shodhana process of *Kaseesa* with different methods

Present study aimed to compare different *Shodhana* methods of *Kaseesa* which is by *Bhavana* in *Bhringaraja Swarasa* (method A), *Swedana* in *Bhringaraja Swarasa* (method B), *Bhavana* in *Nimbu Swarasa* (method C) and *Bhavana* in citric acid solution rather than *Nimbu Swarasa* (method D).

By performing these experiments, it can be quoted, that which method has more challenges for *Shodhana* and which is considerably delightful or convenient to perform. The amount of yield at the end was also compared. Though, in the matter of *Kaseesa, Shodhana* is not the terminal point, but it is an intermediate step, yet, the final yield was considered because of the aspect of standardization.

Method A

In this method, 3 *Bhavana* of *Bhringaraja Swarasa* were given. The method is stated in AFI.[2] Total three (03) of 120g sample size was processed completely dried after *Bhavana* and under sun light.

Table 1: Quantity of *Bhringaraja Swarasa* used in *Bhavana* Process

Batch	Quantity of *Bhringaraja Swarasa* in *Bhavana* (ml.)			Total quantity of *Bhringaraja Swarasa* (ml.)	Average
	1st	2nd	3rd		
A1	30	23	25	78	26
A2	30	25	20	75	25
A3	30	22	25	77	25.6

After *Shodhana* on an average 119.6 g (99.72 %) of yield was obtained from 120 g of *Ashuddha Kaseesa*

Table 2: Quantity of *Kaeesa* before and after *Shodhana* (Method A)

Batch	Quantity of AK (g)	Quantity of BBK (g)	Yield (%)
A1	120	120	100
A2	120	121	100.83
A3	120	118	98.33

Method B

This method is mentioned in *Rasatarngini, Taranga* 21, *Shloka* number 230. In this method, *Kaseesa Shodhana* is mentioned to be performed by following *Swedana* principle using *Bhringaraja Swarasa* as liquid media for the duration of 3 *Ghadi* (72 minutes).[3] As, *Kaseesa* is found soluble in *Bhringaraja Swarasa,* it was retrieved from liquid media by *Bhanupaka* (drying under direct sun light) after completion of *Swedana*.

Table 3: Quantity of *Kaseesa* before and after *Shodhana* & Quantity of *Bhringaraja Swarasa* before and after *Swedana*

S.No.	Parameters	B1	B2	B3	Average
1.	Weight of AK (g)	120	120	120	120
2.	Volume of *Bhringaraja Swarasa* (ml)	1000	1000	1000	1000
3.	*SBK* (g)	135	125	130	130
4.	Percentage of *Shuddha Kasisa* obtained (%)	112.5	104.16	108.33	108.33
5.	Time required for *Swedana* of *Kaseesa* (minutes)	72	72	72	72
6.	Time required for sun drying (days)	3	2	2	2.33
7.	Volume of *Bhringaraja Swarasa* after *Swedana* (ml.)	700	650	630	660

Total 3 batches of 120g sample size were prepared and on average 130 (108.33 %) of yield was obtained by this method. In each *Swedana* process, average 1000ml of *Bhringaraja Swarasa* was utilized.

At the initial phase of *Swedana,* the liquid was added till the bundle got completely immersed into it. After that, the level was maintained during entire process. The average temperature during whole process is 105°C ±5°C.

This method is not widely applied due to unclear explanation of complete details like no explanation about how to recover *Kaseesa* from liquid media. Thus, the attempt to standardize this aspect was covered in present study.

Method C

This method is mentioned in *Bhrihat Rasaraja Sundara,* chapter number 28, *Shloka* number 5-6. Here the principle of *Mardana* is applied by using *Nimbu Swarasa* for 1 day (6 Hrs.) and then it was dried under sun light.[4]

Table 4: Quantity of *Nimbu Swarasa* used in *Maradana* Process

Batch	Quantity of *Nimbu Swarasa* during levigation (ml)							Total quantity of *Nimbu Swarasa* (ml)	Average (ml)
	1st	2nd	3rd	4th	5th	6th	7th		
C1	12	10	8	10	10	10	8	68	9.71
C2	10	8	10	10	6	10	5	59	8.42
C3	10	5	10	8	12	10	6	61	8.71

Total 3 batches of 50g sample size were prepared and on average 48 g (96%) of yield was obtained by this method. On an average 62.66ml of *Nimbu Swarasa* was required to give *Mardana* for one day (6 hrs.).

Table 5: Quantity of *Kaeesa* before and after *Shodhana* (Method C)

Batch	Quantity of AK (g)	Quantity of MNK (g)	Yield (%)
C1	50	48	96
C2	50	46	92
C3	50	50	100

The reason to keep sample size up to 50 g only was to make an additional attempt to modify as stated method D.

Method D

This method is a modified version (*Anubhuta Vidhi*) of method C. Here the entire methodology was exactly the replica of method C, but only difference was in liquid media. In method C, *Nimbu Swarasa* was used while here in method D 6.5% citric acid solution was used as liquid media. The main aim to carry out such experiment was to crosscheck the comparative chemical characteristics of *Nimbu Swarasa* and citric acid solution because citric acid is the main active chemical component of *Nimbu Swarasa.*

Table 6: Quantity of citric acid solution used in *Maradana* Process

Batch	Quantity of Citric acid Solution during levigation (ml)						Total quantity of Citric acid Solution (ml)	Average (ml)
	1st	2nd	3rd	4th	5th	6th		
D1	10	8	10	10	10	2	50	8.3
D2	10	10	8	10	6	10	54	9
D3	10	6	10	8	12	10	56	9.3

Table 7: Quantity of *Kaeesa* before and after *Shodhana* (Method D)

Batch	Quantity of *Asuddha Kaseesa* (g)	Quantity of *Shuddha Kaseesa* (g)	Yield (%)
D1	50	40	80
D2	50	45	90
D3	50	42	82

Total 3 batches of 50g sample size were prepared and on an average 42.33gm (84%) of yield was obtained in this method. Total 53.33 ml of citric acid solution was required for the process.

RESULT

Table 8: Observation and result of *Kaseesa Shodhana* by different methods

Method	Average liquid required in *Bhavana* (ml)	Average liquid required in *Swedana* (ml)	Average duration	Average yield (%)
A	76.66	-	4 days	99.72
B	-	1000	72 min (48 hrs. to dry)	108.33
C	62.66	-	6 hrs. (24 hrs)	96
D	53.33	-	6 hrs. (24 hrs)	84

Analytical study

The analytical study was carried out with a view to know the particular chemical configuration of raw and final products and to check out modifications observed after different *Shodhana* process.

Name of samples

Sample AK: *Ashuddha Kaseesa*

Sample BBK: *Shuddha Kaseesa* (*Bhavana* of *Bhringaraja Swarasa*)

Sample SBK: *Shuddha Kaseesa* (*Swedana* of *Bhringaraja Swarasa*)

Sample MNK: *Shuddha Kaseesa* (*Mardana* of *Nimbu Swarasa*)

Sample MCK: *Shuddha Kaseesa* (*Mardana* with Citric Acid Solution) all the samples of *Ashuddha* and *Shuddha Kaseesa* were analysed using following Parameters:

1. Organoleptic Characters
2. Physico-chemical parameters
   - Determination of moisture content (Loss on drying) [5]
   - Specific gravity[6]
   - Determination of pH values [7]
   - Solubility in water [8]
   - Determination of total ash [9]
   - Determination of acid-insoluble ash [10]
3. Sophisticated instrumental analysis
   - ED-XRF Spectroscopy

Organoleptic characteristics

Table 9: Organoleptic characters of various samples of *Kaseesa*

Sample	Characteristics				
	Nature	**Colour**	**Odour**	**Touch**	**Taste**
AK	Crystalline	Light Greenish	None	Rough and in massive crystal form	Bitter and Metallic
BBK	In powder form	Dark Greenish	Characteristic (fragrant)	Rough and in free-flowing powder form	Bitter and Metallic
SBK	Small lumps and powder	Dark green	Characteristic (fragrant)	Rough and in lumps with powder form	Bitter and Metallic
MNK	In powder form	Light yellowish white (on storage it becomes light Brown	Light lemon fragrance	Smooth and in powder form with hygroscopic tendency	Initially sour then Metallic and Metallic
MCK	In powder form	White	None	Smooth and in powder form with hygroscopic tendency	Metallic

Physico-chemical parameters

Table 10: Physico-chemical parameters performed on *Shuddha* and *Ashuddha Kaseesa*

S.No.	Parameters	Observation of Samples				
		AK	BBK	SBK	MNK	MCK
1.	L.O.D. Loss on Drying (%)	10	65.87	66.70	70.9	71.8
2.	Specific gravity of *Kaseesa Drava*	1.009	1.008	1.007	1.009	1.013
3.	pH of *Kaseesa Drava*	2.8	2.97	3.3	2.22	2.8
4.	Solubility in water (%)	56	53.9	53.6	70.2	83.1
5.	Total Ash (%)	34.62	29.36	29.92	27.69	30.12
6.	Acid Insoluble Ash (%)	12.61	25.70	25.87	26.98	28.85

Table 11: Result of ED-XRF Spectroscopy

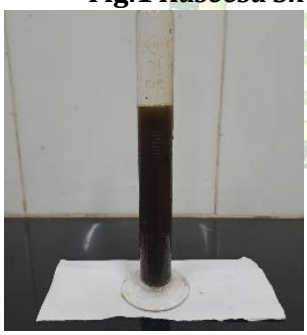
Sample	Total Iron (%)	Total Sulphate (%)	Total inorganic matter (%)
AK	26.03	20.13	48.129
BBK	23.52	17.77	45.777
SBK	21.96	21.24	50.299
MNK	26.6	16.99	45.843
MCK	41.46	31.03	75.404

Fig. 5 ED-XRF Spectroscopy Spectra

XRF Spectra of AK	XRF Spectra of BBK
XRF Spectra of SBK	XRF Spectra of MNK
XRF Spectra of MCK	Overlay spectra of all samples

Fig.1 *Kaseesa Shodhana* (Method A)

Powdered *Ashudha Kaseesa*



Bhringaraja Swarasa was measured

Bhavana of *Bhringaraja Swarasa* for 3 times

Shuddha Kaseesa (BBK)

Fig.2 *Kaseesa Shodhana* (Method B)

Ashudha Kaseesa

Bhringaraja Swarasa was measured

Swedana in *Bhringaraja Swarasa* for 72 min by *Dolayantra*

Removal of *Pottali* and allow it to solidify

Paste spread over steel plate

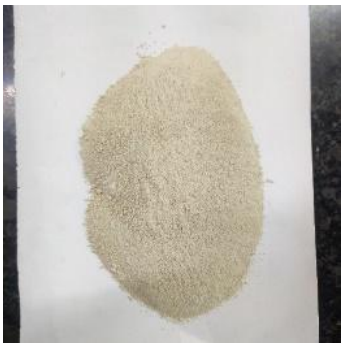
After sun drying *Shuddha Kaseesa* (SBK)

Fig.3 *Kaseesa Shodhana* (Method C)

Powdered *Ashudha Kaseesa*

Nimbu Swarasa was measured

Maradana with *Nimbu Swarasa* for 6 hrs.



Shuddha Kaseesa (MNK)

Fig.4 *Kaseesa Shodhana* (Method D)

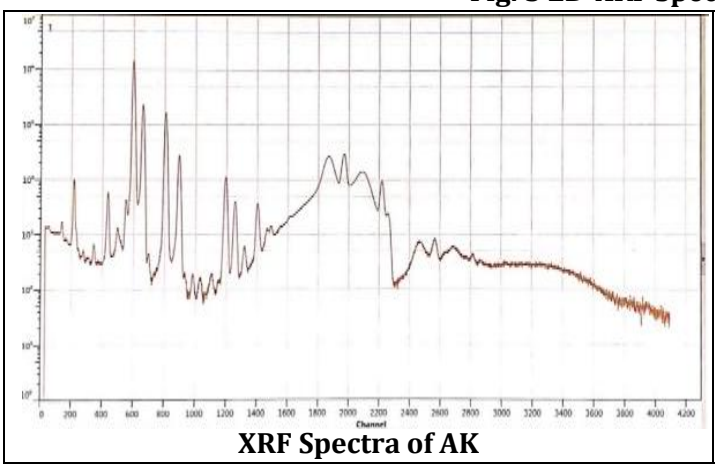
Powdering of *Ashudha Kaseesa*

Citric acid solution was measured

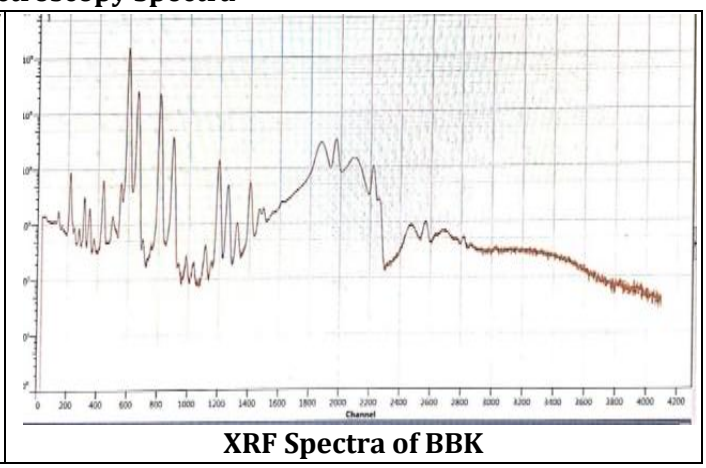
Maradana of Citric acid solution for 6 hrs.

Shuddha Kaseesa (MCK)

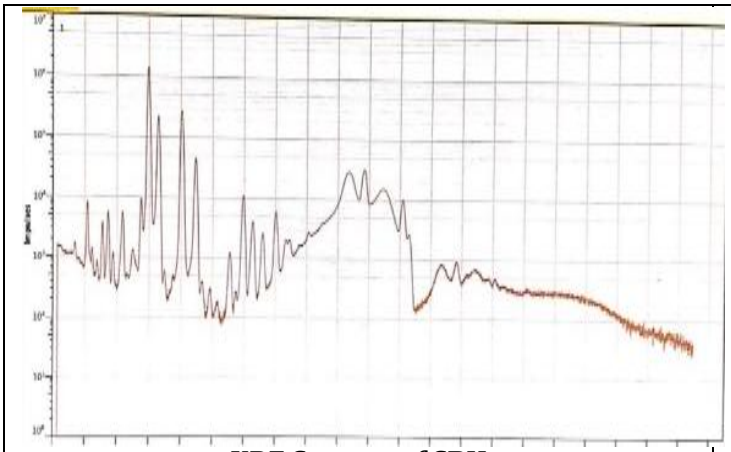
Fig. 5 ED-XRF Spectroscopy Spectra



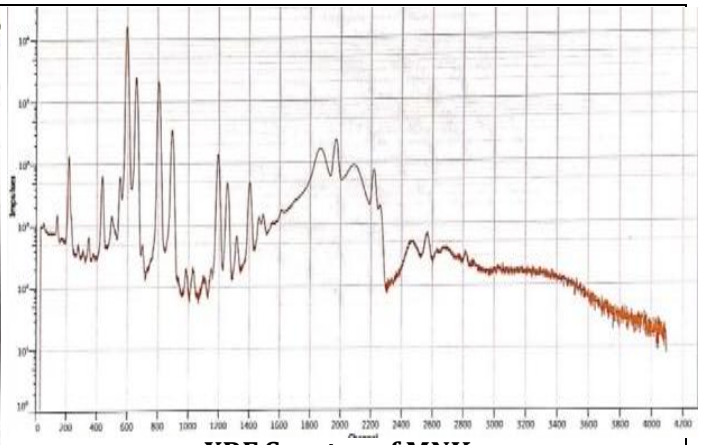
XRF Spectra of AK



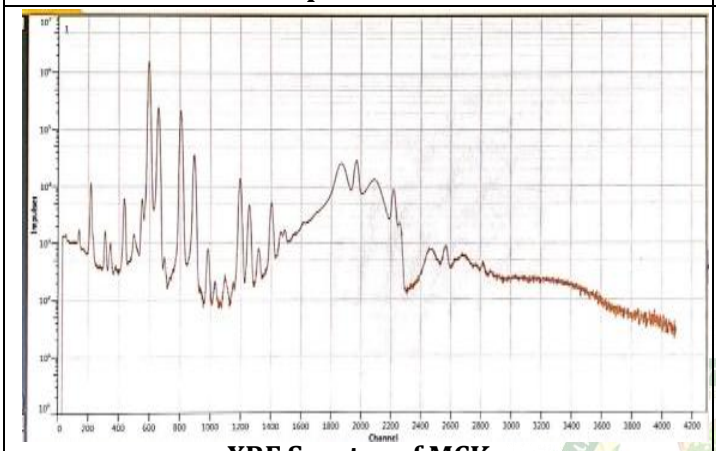
XRF Spectra of BBK



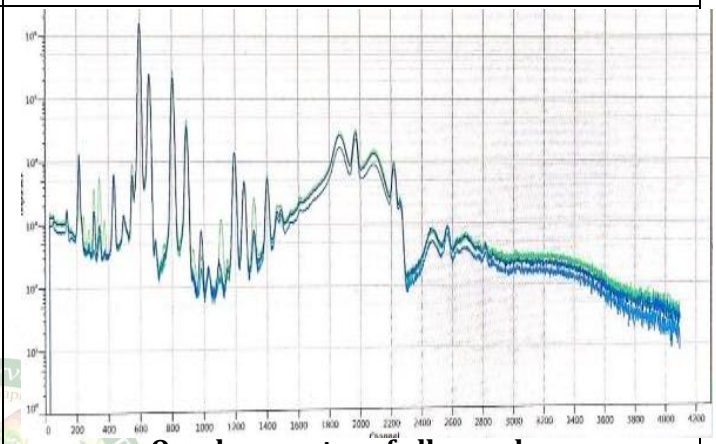
XRF Spectra of SBK



XRF Spectra of MNK



XRF Spectra of MCK



Overlay spectra of all samples

DISCUSSION

Pharmaceutically preparation of BBK, MNK and MCK was much easier and because of its simple mechanism, this method is largely performed at industrial level using the machine called as 'Edge Runner' and/or 'End Runner'. *Swedana* in *Bhringaraja Swarasa* (method D, sample SBK) is not widely applied due to unclear explanation of complete details like no explanation about how to recover *Kaseesa* from liquid media. Thus, the attempt to standardize this aspect was covered in present study. This method is tedious but gives good yield and also removes physical impurities unlike *Bhavana* process. But at larger scale, it is quite challenging to adapt this method. Remarkable variations in organoleptic characteristics were clearly observed in different samples of *Kaseesa* that was undergone different method of *Shodhana* and using different liquid media.

Moisture content can be determined by performing L.O.D. Above table 10 shows that initially moisture is present in the raw form in sample AK but after *Shodhana* it has high amount of moisture. Although in its chemical formula 7 molecules of water are present, *Kaseesa* (ferrous sulphate) is hygroscopic in nature and after *Shodhana* it becomes more hygroscopic. Sample MNK and MCK are having higher amount of moisture content. Thus, it would be preferable to store it in an air tight container. Specific gravity of *Kaseesa Drava* shows the presence of the amount of dissolved compound in water. Sample AK and MNK are having same concentration. While sample MCK is having highest concentration among all. Sample BBK and SBK are having lowest concentration. This indicates more dissolution may have been happened in sample MCK. pH of *Kaseesa Drava* indicates either acidic or basic nature of solution. Usually, ferrous sulphate is found acidic in nature. pH of *Kaseesa* that has undergone *Shodhana* with *Bhringaraja Swarasa,* was found to be more basic than *Ashuddha Kaseesa.* Probability of this changes could be the presence of *Bhringaraja Swarasa* particles. pH of *Kaseesa* that has undergone *Shodhana* with *Nimbu Swarasa* and citric acid, was found acidic. This could be because of acidic liquid media. The reading of pH shows that, though average range of pH of all samples is acidic, yet the process of *Shodhana* does affect the pH of *Kaseesa.*

Solubility of *Kaseesa* as above mentioned in table 10 was increased in sample MNK and MCK when it was compared with the raw sample. While it was found to be decreased in sample BBK and SBK

when it was compared with the raw sample. Ash value or total ash is 34.62%, 29.36%, 29.92%, 27.69% and 30.12% of sample AK, BBK, SBK, MNK and MCK respectively. Which indicates that raw *Kaseesa* has high total Ash values than *Shuddha Kaseesa.* Acid insoluble ash is 12.61%, 25.70%, 25.87%, 26.98%, and 28.85% of sample AK, BBK, SBK, MNK and MCK respectively. Which indicates the presence of inorganic matter was found high in *Shuddha Kaseesa* when compared to *Ashuddha* one. The above table 11 shows that the AK which is *Ashuddha Kaseesa* as initial raw material has 26.03 % of iron, 20.13% sulphate and total 48.129% of inorganic matters.

In BBK, the amounts of iron, sulphate and total inorganic concentration are found decreased (23.52% iron, 17.77% sulphate and total 45.777% of inorganic matters) when compared with Sample A. The probable reason of this decreased amount is the addition of organic matter. In SBK**,** iron concentration (21.96%) and sulphate (21.24%) are found decreased while total inorganic concentration was found increased (50.299%). Remarkable decrease in iron content could be because of addition of organic matter. Whereas, increase in total inorganic salts shows chances of higher amount of inorganic matter in *Bhringaraja*. MNK show slight increase in iron concentration (26.6 %). But sulphate (16.99%) and total concentration of inorganic matter (45.843%) decreased. This could be because of chemical reaction between *Kaseesa* and *Nimbu Swarasa.*MCK shows that iron, sulphate and total concentration of inorganic matter has been drastically increased which is 41.46%, 31.03% and 75.404%. Here its density was also found increased. Which could be because of probable chemical reaction between *Kaseesa* and citric acid solution.

CONCLUSION

Pharmceutico-analytical study revealed that there were major physical and chemical changes occurs after *Shodhana* in *Kaseesa*. Each method and liquid media led to major physical and chemical changes in *Kaseesa*. Total iron content in *Ashuddha Kaseesa* (sample AK) was 26.03% which was compared with *Shuddha Kaseesa* samples process with different methods and medias. Three *Bhavana* of *Bhringaraja Swarasa* is a simple method (method A, sample BBK). It yields an average more than 99%. Compare to other samples, it is more stable but total iron content was decreased which is 23.52% as compared to AK. *Swedana* in *Bhringaraja Swarasa* for 3 *Ghadi* (72 minutes) method (method B, sample SBK) was tedious but found to be more affective in removal of physical impurities. The method yields an average of 108.33%. Compare to other samples it is more stable but total iron content was decreased which is 21.96% as compared to AK. *Mardana* with *Nimbu Swarasa* is simple method (method C, sample MNK). It yields an average more than 96%. Compare to other samples it is hygroscopic and gives colour change during storage but total iron content was increased slightly which is 26.6 % as compared to AK. *Mardana* with citric acid solution is *Anubhut* or modified method (method D, sample MCK). It yields an average more than 84%. Compare to other samples it is hygroscopic and gives colour change during storage but total iron content was increased which is 41.46% as compared to AK.

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***Address for correspondence**
Ms. Bhanderi Radhika
M. Pharm (Ayu) Scholar,
Department of Rasashastra and Bhaishajya Kalpana, Institute of Teaching and Research in Ayurveda, Jamnagar.
Email: radhi24bhanderi@gmail.com