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PHYSICO-CHEMICAL ANALYSIS AND EVALUATION OF ANTIBACTERIAL AND ANTIFUNGAL ACTIVITY OF *KITMARDO RASA*

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Abstract

Ayurveda as an ancient science developed through trials, experiments, and observations. Ayurveda has been honored and applied for the prevention and cure of human ailments with purity and sacredness. Ayurvedic medicines have proven to be effective in treating various diseases using metallic, non-metallic, and compound materials. The segment of RASASHASTRA focuses on inorganic pharmaceutical preparations, which require the process of SHODHANA to make toxic raw materials safe for consumption. Different types of Shodhana techniques are described in RASAGRANTHAS to convert raw materials into effective medicinal formulations. Techniques like Marana, Parpati Kalpana, KupipakwaRasaayans, and Kharlya Kalpana are used to prepare medicines based on ancient Granthas for curing ailments. The field of RASASHASTRA deals with mercury, minerals, metals, and aquatic substances categorized based on their actions on Dhatu and the body. Kajjali, a combination of Parada and Gandhaka, is widely used in formulations and has therapeutic applications on its own. Kajjali is indicated for various conditions like Visarpa, Vidhradhi, Charmadal, Kandu, Pama, and Upadandha, which are correlated with infectious diseases. The abstract also emphasizes the need to find new antimicrobial compounds due to the development of antibiotic-resistant bacteria. Ayurvedic herbo-mineral preparations are suggested as potential sources for antimicrobial activity evaluation. The Kitmardo Rasa, a combination of various ingredients, is discussed as a Karliya Kalpana formulation, and the study aims to evaluate its physico-chemical properties and antibacterial/antifungal activity in vitro.

Introduction

"*Ayurveda*" is the ancient science, which was developed by the various stages, by way of continues trials, experiments and deliberate minute observations. However, *Ayurveda* is ancient Science, it has been honored, respected and applied for the prevention and cure of human beings with purity and sacred. The *Ayurvedic* Medicines are proved to be more officious in various diseases. The segment of the *RASASHASTRA*, covers the exclusive studies and experiments of inorganic Pharmaceutical Preparations such as *AyurvedicRasaushadhis* plays very vital role in to improve the scope of *Ayurveda.*

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The Ancient Indian alchemy is dealing with *PARADA* (Mercury) i.e. *RASA*, Minerals, Metals and aquatic substances, all are generally considered in *RASASHASTRA*. *Kajjali* which is a combination of *Parada* and *Gandhaka*, forms the basis for most *Kharaleeya*, *KupiPakva*, *Pottali* and *ParpatiKalpanas*. It is used in many of the formulations and has numerous therapeutic applications on its own.

Pathogenic microorganisms are responsible for many life-threatening diseases. Antibiotics are used successfully to treat these infectious diseases. But indiscriminate use of antibiotics has developed many resistant strains of bacteria not responding to any currently available antibiotic. So, there is need to search new antimicrobial compounds. *Ayurvediya*herbo-mineral preparations are used according to Rasa, *Guna*, *Virya*, *Vipaka*, *Prabhava* etc. The scope to evaluate antimicrobial activity of these compounds can be extended. The purpose of the present study was to evaluate scientifically the "Physico-chemical analysis and evaluation of antibacterial and antifungal activity of *KitmardoRasa*" in vetro.

Aim and Objectives

Aim:

- To establish the physico-chemical standards for *Kitmardo Rasa.*
- To assess the antibacterial and antifungal activity of *Kitmardo Rasa.*

Objectives:

- To prepare *Kitmardo Rasa* as per *Rasendrasarsangraha.*
- To conduct Physico-chemical analysis of*Kitmardo Rasa.*
- To conduct in vitro evaluation of Antibacterial and Antifungal activity of *Kitmardo Rasa.*

Material and Methods

Materials:

1. Sample of raw Parada, raw Gandhaka, saindhavaLavana, Sudha, Godugdha and Goghruta were collected from our college Pharmacy.
2. Sample of Rason, Kuchala, Ajamoda, Vidanga, Palasha was collected from our college Pharmacy.

Methods:

This chapter elaborates methods in three Parts as-

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1. Pharmaceutical study
2. Analytical study
3. Antimicrobial study

Result and Discussion

Utilization of elements to relieve man and animals from disorders, as well as for maintenance of healthy status, is as old as life on earth. Ayurveda is the oldest science. It emphasizes on maintenance of health and cure of disease. For achievement of these goals, medicines especially Rasaushadhies play an important role. Question regarding the toxicity of mercury arise again and again after certain period. So the present study is an attempt to evaluate the potency of' KajjaliyuktaKharaliyakalpana' which is a mercury based compound.

Kajjali forms the base for the various kalpanas viz., kharaliya, Parpati, Kupipakwa and Pottali. The properties of kajjali vary in each kalpana depending on the Samskar done on it and the quality and quantity of agni to which it is subjected. This can be explained by the simple example seen in our day- to- day life. From the same wheat flour we can prepare chapatis, puris or bread with change in preparation method and heat modality (frying, roasting, puffing etc.) The efficacy of Aushadhis prepared using the kajjali depend on the quality of the kajjali used. Thus, it is essential to have good & complete knowledge of kajjali.

The discussion of present study is divided into three parts i.e. Pharmaceutical study, Analytical study, antimicrobial study.

Pharmaceutical Study:

Parada Shodhan:-

I. Sudha Shodan:-

Sudha Shodhan was done according to the text Rasatarangini. When Sudha churna was mixed with water it easily mixed with it and colour changed to muddy white. When the mixture was filtered, the impurities settled on cloth piece, thus Shodhan brightens colour. The time required for sedimentation was 7 hrs. While decanting precaution should be taken to avoid mixing of Sudha with water.

According to reference quoted in Rasatarignani. Parada shodhan was done. The duration of trituration is mentioned as a मर्दयेत्र्यहम्, for three days so 12 hrs per day i.e. 12 × 3 = 36 hrs, was decided for Parada shodhan.

Trituration was done for 4-5 hrs/day. Parada was get collected in the middle of khalva which was shining. This Parada was collected in the glass jar separately. Remaining Sudha was washed with hot water and remaining Parada was collected after vastragalana.

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At the time washing Parada settled at the bottom of vessels which was easily collected. Some fine particle float on the surface of water which get washed away with water as explained in text as Jalagati and Hansagati of Parada. So care was taken to avoid loss of Parada. After Shodhan shine of Parada increased.

Sr. No.	Weight of Parad Before Shodhan in sudha	Weight of Parad after Shodhan in sudha	% loss
1.	100 gm	98 gm	2%

II. Parada shodhan with Rason and SaindhavaLavan:-

According to reference same as above quoted in Rasatarignani 5/27-30 Parada shodhan was done. “so mardana is to be done till rason Kalka turned into Krushna varna. After 60 hrs of MardanRasona Kalka turned into blackish Green. Trituration was done for 4-5 hrs/day. Parada was washed with hot water and remaining Parada was collected after vastragalana. Parada was collected in the glass jar separately.

At the time washing Parada settled at the bottom of vessels which was easily collected. Some fine particle float on the surface of water which get washed away with water as explained in text as Jalagati and Hansagati of Parada. So care was taken to avoided loss of Parada. After Shodhan shine of Parada increased.

Role of ShodhanaDravya:-

It is noted that the shodhanadravya are selected according to their particular action in the shodhana process. They are easily available. Parada has Naisargika, yogika and aupadheyikadosha. This trituration with shodhanadravya might alter the dosha of Parada and make it biological acceptable form.

Changes of Material during Mardana process

1. Physical changes

   In Maradana process Parada become more shiny.
2. Chemical changes

   In Maradana process chemical reactivity of Parada increases.

Sr. No.	Weight of Parad Before Shodhan in Rason and saindhav	Weight of Parad after Shodhan in Rason and saindhav	% loss
1.	98 gm	85 gm	13.265%

- **GandhakShodhan :-**

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According to reference quoted in RasatarignaniTarang 8/7-12 Gandhakshodhan was done. Gandhaka was powdered finely in khalwa yantra. This powdered Gandhaka was taken in a lohadarvi, smeared with Goghrita and then subjected to mandagni. Color changed from Dark yellow to oranges yellow during liquefaction of Gandhaka. Gandhaka was stirred with spoon intermittently. When Gandhaka melted completely, it was poured into a vessel containing Godugdha. The Gandhaka from Godugdha was collected and washed thoroughly with hot water.

This procedure was repeated for 3 times. Ugragandha of Gandhaka reduced considerably in subsequent procedures. In the end of the procedure color of Godugdha turned to yellowish white with oily appearance. After each process Gandhaka became more brittle.

- **Mode of Action of Sodhana Process of Gandhak:-**

  According to commentary by D.A.Kulkarni on Rasaratnasmucchya. The substances used for GandhakaSodhana are Ghruta, Godugdha, Agni, Vashtra. The use of all these are interrelated with each other. Mainly the impurities present in Gandhaka are follows.

  i. Impurities like stone and other extraneous materials which are present in Gandhaka, they get separated when Gandhaka is heated and melted.

  ii. Some impurities get melted and dissolve in Ghruta and milk and are separated.

According to above reference quoted in RasatarignaniTarang 2/27-28 Prepartion of dwigunaKajjali was done. While preparation of KajjaliDrudhaMardana is carried.

Shodhit Parada and its double quantity of ShodhitGandhak were taken in khalva yantra and triturated continuously for 8 hrs/day till Kajjali became Nischandrika. As trituration started parada globules commence to convert into fine practicals. Gradually the color of mixture changed and luster of mixture diminished, but chandrikas were present. After the completion of trituration black colored lusterless powder was formed. Metallic luster is called as chandrika.

After 50 hrs of mardanakajjali became Nischandrika. A small quantity of prepared kajjali was taken in between thumb and index finger and rubbed vigorously (Rekhapurnatva). The portion of powder adhered into the grooves of these fingers was exposed to the sunlight and viewed carefully (Nischandrikatva). Absence of any tiny shiny particle indicates proper formation of kajjali.

Sr. No.	Weight of ShodhitParad	Weight of ShodhitGandhak	Expected wt of Kajjali	Wt. Of acquired Kajjali	% loss

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1.	40 gm	80 gm	120 gm	114 gm	5%

- ***Kuchala*Shodhan -**

  According to *Rasendrasarasangraha.* In this process 300 gm *AshudhaKuchala*was taken for *Shodhana* it was observed that 20gm loss took place in this process.

Sr. No.	Weight of *Kuchala* Before Shodhan	Weight of *Kuchala*after Shodhan	% loss
1.	300 gms	280 gms	6.66%

- ***AjamodaChurna*preparation :**

  In this process 200 gms*AjmodaChurna*was taken for preparation. It was observed that 10 gms loss took place. Total obtain amount 190 gms in this process the average presentag loss is 5%.

Sr. No.	Total Qty. of *AjamodaChurna*taken	Total Qty. of *AjamodaChurna*obtained	Total weight loss	% loss
1.	200 gms	190 gms	10 gms	5%

- ***VidangaChurna*preparation :**

  In this process 200 gms*VidangaChurna*was taken for preparation. It was observed that 15 gms loss took place. Total obtain amount 185 gms in this process the average presentag loss is 7.5%.

Sr. No.	Total Qty. of *VidangaChurna*taken	Total Qty. of *VidangaChurna*obtained	Total weight loss	% loss
1.	200 gms	185 gms	15 gms	7.5%

- ***PalashaChurna*preparation :**

  In this process 300 gms*PalashaChurna*was taken for preparation. It was observed that 10 gms loss took place. Total obtain amount 290 gms in this process the average presentag loss is 3.3%.

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Sr. No.	Total Qty. of *PalashaChurna*taken	Total Qty. of *PalashaChurna*obtained	Total weight loss	% loss
1.	300 gms	290 gms	10 gms	3.3%

Peparation of Kitmardo rasa:

- Kitmardo rasa had prepared by' KajjaliyuktaKharaliya Kalpana' method given by Rasendrasarasangraha.
- Observation presented in tabular and charts format in pharmaceutical study in shorts observations are given below

Time and date while preparing of

'Kitmardo Rasa'

Start Date and Time	End Date and Time	Total Amount Taken	Total obtained Amount	Total Weight loss	Loss of % duration preparation
25/08/15 06:00 am	25/08/15 08:00 pm	840 gms	800 gms	40 gms	5%

AnalyticalStudy:

Analytical study is determination of the values of different parameters used for the analysis of a sample. It brings standards for the quality drug and helps in explaining pharmacokinetics and pharmacodynamics of a drug. Analytical study was carried out with a view to know particular chemical configuration of the final product and to point out the physico-chemical profiles of the drugs. It is the marking line to note the limits or ranges of the values. Hence, it stands as a method of standardization of formulations. For standardization of finished product it is essential to analyze the prepared drug by employing various parameters and then to fix suitable standards, so that quality of the product can be established.

In this phase, analytical study of raw material, in process and finished products were done by employing various parameters. The parameters adopted are macroscopic characters, physicochemical parameters, chromatographic study and Antimicrobial study.

i. Owing to removal of physical impurities colour of Sudha turned from dull white to bright white.

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ii. The Shodhit Parada follows the ayurvedic norms of grahyaparada. Also the % purity of Parada after Shodhan was observed to be 98.64% and that of Ashodhit Parada being 97.30%. Purity may be increased due to the chemical reaction with Sudha and Rasona because of Maradana and repeated Dahvana and Galana processes.

iii. The ShodhitGandhaka follows the ayurvedic norms of grahyaGandhaka. Also the % purity of Gandhaka after Shodhan was observed to be 98.08% and that of AshodhitGandhaka being 97.50%. Purity may be increased due to ShodhanSanskar and repeated Dhalan, Galan, Dahvana processes. There was change in bulk density of GandhakaAshodhit and Shodhit, they were 1.9381 gm/ml and 1.9401 gm/ml respectively. There was change in Melting point of GandhakaAshodhit and Shodhit, they were 116.8^0 C and 118^0 C respectively.

iv. Preparation ofDwigunaKajjali contains % of Mercury 32.58% respectively. There was change in Total sulphur content of DwigunaKajjali, they were 48.73% respectively.

Physico-chemical Analysis of Kajjali

Sr. No.	**Tests Names**	**DwigunaKajjali**
1	Description	A Black colour fine powder prepared from mercury &sulphur
2	Feel	Soft
3	Moisture content	0.83%
4	Mercury content	32.58%
5	sulphur content	48.73%
6	Ash content	0.92%
7	Water insoluble ash	<0.1%
8	Acid insoluble ash	<0.1%

- **Physico-chemical Analysis of VidangaChurna:**

SSr. No.	**Parameters**	**VidangaChurna**
1	Appearance	Slightly Redish Color Moderately

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		fine powder
2	Odor/Test	Slightly Aromatic/Astringent
3	Moisture Content	5.49%
4	pH	6.55%
5	Ash content	3.97%
6	Acid insoluble ash	0.30%
7	Water Soluable Extractive	29.24%
8	Alcohol Soluable Extractive	16.72%

- **Physico-chemical Analysis of PalashaChurna:**

Sr. No.	Parameters	PalashaChurna
1	Appearance	Slightly Yellowish brown Color Moderately fine powder
2	Odor/Test	Faint/Slightly astringent
3	Moisture Content	5.76%
4	pH	7.04%
5	Ash content	8.46%
6	Acid insoluble ash	1.43%
7	Water Soluable Extractive	30.71%
8	Alcohol Soluable Extractive	18.66%

a. Physico-chemical Analysis of AshodhitKuchala and ShodhitKuchala :

Sr.No.	Parameters	AshodhitKuchala	ShodhitKuchala
1	pH	5.89	6.14
2	Moisture Content	5.64%	8.78%
3	Total Ash content	1.42%	1.22%
4	Acid insoluble ash	< 0.1%	< 0.1%

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5	Water Soluble Extractive	29.60%	31.10%
6	Alokohol Soluble Extractive	13.79%	15.60%

- **Physico-chemical Analysis of AjamodaChurna :**

Sr. No.	Parameters	AjamodaChurna
1	Appearance	Slightly Yellowish Brown Color Moderately fine powder
2	Odor/Test	Faint/Slightly astringent
3	Moisture Content	5.76%
4	pH	7.04
5	Ash content	8.46%
6	Acid insoluble ash	1.43%
7	Water Soluable Extractive	30.71%
8	Alcohol Soluable Extractive	18.66%

6.4 Antibacterial and Antifungal Study:-

From Antibacterial and Antifungal Study using Serial dilution and Disc diffusion Method was concluded.

- Antibacterial activity of Kitmardo Rasa is less significant with compared to standard (Benzthin penicillin).
- Antifungal activity of Kitmardo Rasa is highly significant with compared to standard (Flucanzol).
- Form this present study it may be concluded that the Antifungal activity of Kitmardo Rasa is found to be better than Antibacterial activity during in Vitro study.

Analytical table of Antibacterial activity:-

Micro-organisms	MIC (Serial dilution method)	Zone of Inhibition (Disc diffusion method)	Comparative Analysis

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	Stan dard	**Sample Drug**	**Stan dard**	**Sample Drug**	**Standard**
Streptococci Shigile	2µg/ ml	25µg/ml	20 mm	16 mm	+
E.coli	2µg /ml	100µg/ml	20 mm	13 mm	+

Indication: - + Better activity .

Analytical table of Antifungal activity: -

Micro-organisms	**MIC (Serial dilution method)**		**Zone of Inhibition (Disc diffusion method)**		**Analysis**
	Standard	Sample Drug	Standard	Sample Drug	Sample Drug
Candida albicans	2µg /ml	0.2µg/ml	20 mm	25 mm	+
Aspergillus niger	2µg /ml	0.2µg/ml	20 mm	24 mm	+
Trichophyton rubrum	2µg /ml	0.2µg/ml	20 mm	24 mm	+
Cryptococcus neoformans	2µg /ml	0.2µg/ml	20 mm	24 mm	+

Indication: - + Better activity than other.

So, from Antibacterial and Antifungal study conclusion can be drawn as Kitmardo Rasa Shows better antifungal activity than that of antibacterial.

Summary

The dissertation topic entitled "physico-chemical analysis and evaluation of antibacterial and antifungal activity of Ktimardo Rasa composed by 8 sections i.e.

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Introduction, Aim and Objectives, Review of Literatures, Materials and Methods, Observations and Results, Discussion, Summary, Conclusion.

1. Brief introduction is given in the beginning of dissertation, which depicts importance of Rasashatra in Indigenous System of Medicine and also justification of present research work.
2. Aim and Objectives are mentioned in this section.
3. Review of Literature includes 9 subdivisions.
    a. It deals with description conceptual study of Murcchana with its different types.
    b. It elaborates information about Parada which includes historical review, synonyms, types, Shodhana process, therapeutic uses, AmayikaProyoga, Dose, Anupana etc. Ayurvedic and modern view.
    c. It elaborates information about Gandhaka which includes historical review, synonyms, types, Shodhana process, therapeutic uses, AmayikaProyoga, Dose, Anupana etc. Ayurvedic and modern view.
    d. It deals with description of conceptual study of Shodhana with its different types.
    e. It deals with description of shodhanaDravya that is Sudha, SaindhavaLavana, Lashuna, Godhudha, Goghruta. Its Ayurvedic and modern view.
    f. Detail information about synonyms, Kajjali Bandha, Method of preparation, therapeutic uses, amayikaproyoga, dose, anupana of Kajjali is given.
    Ayurvedic and modern view.
    g. It deals with Historical review of concept of microbes in Ayurvedic aspect, brief description about the methods of antibacterial and antifungal study, information about the test organisms etc is mentioned in this.
    h. Review of previous work done related to Kitmardo Rasa and Antibacterial and Antifungal activity has been mentioned.
4. The forth section, Material and Methods, consists of three parts.
    a. Pharmaceutical study deals with practical work performed. The raw material like Parada, Gandhaka, and Shodhandravya Sudha, SaindhavaLavana, Lashuna, Godhudha, Goghruta, Ajmoda, Vidanga, Palasha, Kuchala were collected from our college Pharmacy. All collected raw material were

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authenticated form expert in this field Rasashatra&Bhaisajyakalpana, Dravyaguna. Pharmaceutical study was carried out in Practical No-1 to Practical No-10.

b. It deals with Qualitative and Quantitative analysis of raw drugs before and after Shodhana. Methods of analysis Parameters are mentioned in detail.

c. Antibacterial and Antifungal study of Kitmardo Rasa is described in this part. The present study was the modest attempt in direction of Antibacterial and Antifungal activity of Ayurevediya Formulations. It was done by two methods i.e. Serial dilution method and Disc diffusion method.

5. The fifth section, Observation and Result, consist of Observation and Result in three parts. The results obtained from the observations during Pharmaceutical, Analytical, Antibacterial and Antifungal Studies are mentioned in this section. Results of Organoleptic changes, Melting Point, Bulk Density, % of Sulfur, Total sulfur content, Free Sulfur, % of Mercury by A.A.S., XRD, Ash Content, Moisture Content, Acid insoluble Ash, Water insoluble Ash. Are mentioned in it. Result of the Antibacterial and Antifungal activity of drug was interpreted on the basis of obtained zone of growth inhibition of that particular organism. The sensitivity of these organisms was found to be different at the various concentrations of the test drug samples.
6. The findings observed are critically analyzed and discussed along with the reasoning to draw some fruitful conclusion regarding the topic.
7. It contains Conclusion of this study as given bellow.

   In DwigunaKajjali 40 gms of Parada and its double 80 gms of Gandhaka was taken so it was suppose to be 120 gms of kajjali but I got 114 gms of Kajjali Prepared. Total loss 6 gms, due to Mardana procedure. To preparation of DwigunaKajjali 7 days is required.

   In Kuchalashodhan 300 gms. Of kuchala was taken, I got 280 gms. Total loss was 20 gms. Form this present study it may be concluded that the Antifungal activity of Kitmardo Rasa is found to be better than Antibacterial activity during in Vitro study.

Conclusion

1. **PHARMACEUTICAL STUDY:**

Shodhana of Parada: -

Parada Shodhan with Sudha by Mardanaprocedure: -

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SamyakShodhit Parada Lakshan is observed after 30 hrs.

Parada Maradana with Sudha color of Parada is Silver shiny and turned in to Silver shiny^{+++}. Color of Shodhit Sudha White and turned in to Gray.

Parada shodhan with Rason and Saindhivalavan:-

SamyakShodhit Parada Lakshan is observed after 60 hrs.

Parada Maradana with Rason and Saindhivalavan color of Parada is Silver shiny and turned in to Silver shiny^{+++}. Color of Rason Kalka is yellowish and turned in to Blackish Green^{++}.

The above procedures of Parada Shodhana by triturating in Sudha, RasonaKalKa and SaindhavaLavan are much time consuming.

Shodhan of Gandhaka:-

For Proper GandhakaShodhanaDhalana and Galana Procedures should be repeated for three times. And it is necessary to change Goghruta, Godugdha, for each cycle.

Preparation of Kajjali:

Preparation of KajjaliMardanaSanskara plays an important role. Proper pressure while preparing Kajjali is the Mandatory step. DwigunaKajjali Show Nischandratva in 50 hrs.

Shodhana of Kuchala :

pH of AshodhitKuchala was 5.89% and that of shodhitKuchala was 6.14% thus it could be concluded that during shodhana process pH of shankha increases.

Preparation of Kitmardo rasa:

Preparation of Kitmardo rasa was done by Kharaliyakalpana method mentioned in Rasendrasarasangraha, 2/11-12.

ANALYTICAL STUDY:

From Analytical study it may be concluded that.

- Shodhana of Parada % Mercury increases and also there is increase in brightness of Parada.
- Shodhana of Gandhaka may increases % of Sulfur, bulk density and Melting Point.
- By the physicochemical analysis the following results are obtained Kitmardo rasa is a medicated preparation of soft feel and slight blackish grey colored fine powder.
- It has moisture content 2.43%, total ash content 9.83%, acid insoluble ash 1.07% water soluble ash greater than 18.90%.

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- As it is Paradmaritbhasma so 1.68% mercury content is present on it.
- In this kalpa free sulpher content 5.73%

- **ATOMIC ABSORPTION SPECTROPHOTOMETRY (AAS) AND X-RAY DIFFRACTION (XRD) STUDY:**

Result of Kitmardo Rasa by AAS study:

- Mercury content in Kitmardo rasa is 1.68%
- Sulfur content in Kitmardo rasa is 5.73%
- pH content in Kitmardo rasa is 6.46%

Results of Kitmardo rasa by XRD study:

In XRD analysis, for sample of **Kitmardo Rasa** made using Kajjali& specified herbal material in proper proportion, from the x-ray diffraction pattern it is seen that the phases observed in sample is identical with that of mercuric sulfide to some extent and some peaks are unidentical. The x-ray diffraction pattern is of mixed phase, it indicates the majority presence of mercuric sulfide; however there is some signature presence of sulfur phase to some extent.

2. Antibacterial and Antifungal Study:-

Antibacterial activity:-

1. In serial dilution method, the antibacterial activity of Kitmardo Rasa against Streptococci shigile was equal but was less efficient than the standard.
2. In disc diffusion method the antibacterial activity against Streptococci shigile, of Kitmardo Rasa had less efficacy than the standard.
3. In serial dilution method, the antibacterial activity of Kitmardo Rasa against E.coli was equal but was less efficient than the standard
4. In disc diffusion method the antibacterial activity against E.coli, of Kitmardo Rasa had less efficacy than the standard.

Antifungal activity:-

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1. In serial dilution method, the antifungal activity against Candida albicans, Aspergillus niger, Trichophyto rubrum and Cryptococcus neoformans of Kitmardo Rasa were equal and were better than the standard.
2. In disc diffussion method, the antifungal activity against Candida albicans, Aspergillus niger, Trichophyto rubrum and Cryptococcus neoformans of Kitmardo Rasa had better efficacy than the standard.

So from Antibacterial and Antifungal study conclusion can be drawn as Kitmardo Rasa shows better Antifungal activity than that of Antibacterial.

4. SCOPE FOR FURTHER STUDY :

As Kitmardo Rasa shows good anti-fungal activity in in-vitro study there is further scope for in-vivo study.

Confirmation of Anti-fungal activity of Kitmardo Rasa can be authenticated by animal and clinical study.

Animal and Clinical study can be conducted for fungal infections.

Higher fungal strains can be tried for confirmation of anti-fungal activity of Kitmardo Rasa.

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