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Research Article

PHYSICO-CHEMICAL AND ANTIMICROBIAL STUDY OF *RASAKARPURA DRAVA*.

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ABSTRACT:

Among *Rasaushadhis, Kupipakva rasayanas* hold the top place. The effects of these *Kupipakva rasayanas* are really a miracle. Many *Kupipakva rasayanas* are explained in classics such as *Rasasindur, Rasakarpoor, Sameerpannagarasa, Talasindhur* etc. *Kupipakva rasayanas* are classified into two type's viz. *Sagandha and Nirgandha*. *Rasakarpura* is *Nirgandha Kupipakva rasayana*. There are very less literature and studies are available regarding *Nirgandha Murchhana*. But the *Kupipakva rasayanas* prepared by *Nirgandha Murchhana* are very effective at very low dose. According to *Rasatarangini Rasakarpoor* is having a property *krimigna*, *bahubhootavishapaha*, and *sarvarogahara*. Here an attempt has been made regarding *Rasakarpura* in its liquid dosage form as "*Rasakarpura Drava"* to assess its antimicrobial activity. It was used in two forms water media and Hydro-alcoholic media. An antimicrobial study carried out showed that the products are highly effective when used against *S. aureus, C. albican* and *E. coli.* These observations suggests a new path for the development of user friendly tropical application of *Rasakarpura drava* in various kinds of skin infections and day to day life product like hand wash, instrument sterilizer etc.

KEY WORDS: *Kupipakva Rasayana, Rasakarpura Drava,* Antimicrobial, *Nirgandha Murchhana.*

INTRODUCTION:

Ayurveda is not a sudden invention but gradual evolution. It is not just a curative medicine, but also it teaches the way to live long a healthy and happy life. The imperishable fundamentals of Ayurveda are still applicable because of their scientific eternal background. Such fundamentals of ayurveda must be subjected to scientific research not only to prove its certainty but also to add something to the existing knowledge.

Among *Rasaushadhis, Kupipakva rasayanas* hold the top place. The effects of these *Kupipakva rasayanas* are really a miracle. Their efficacy is good if they are prepared by proper procedures. In the preparation of *Kupipakva rasayana, agni* is an important factor, which changes the natural physico – chemical properties of the drug. Many *Kupipakva rasayanas* are explained in classics such as *Rasasindur, Rasakarpoor, Sameerpannagarasa, Talasindhur* etc. *Kupipakva rasayanas* are classified into two types viz. *Sagandha and Nirgandha*. *Rasakarpura* is a *Kupipakva rasayana*, which is said to be of *Nirgandha* type i.e. during the preparation of *Rasakarpura, Gandhaka* (Sulphur) will not be used directly. Few of the authors have recommended utility of *Gandhakamla* (Sulphuric acid) in the process of *Rasakarpura*.

According to *Rasatarangini Rasakarpoor* is having a property *krimigna*, *bahubhootavishapaha*,

atisar, pravahika, tvachagatarog, raktadoshamana, grahi, spota,kandu, mandala, phiranga, kushta and vranashana and mentioned it as *sarvarogahara*. Considering above all the properties we can assess *Rasakarpura* is one of the *Rasaushadh* which has krimighna property. *Krimi and bahubhootvisha* of Ayurveda are correlated with microorganisms. The *krimis* are explained under two broad headings as visible and invisible in our *Vedas*. According to the recent authors of Ayurveda *bhootas* are those diseases causing organisms, which cannot be seen through the naked eyes.

AIM & OBJECTIVES:

AIM:

Physico-chemical analysis and antimicrobial study of *Rasakarpura Drava*,

OBJECTIVES:

Preparation of *Rasakarpura drava* with water (Aqueous) media by following procedure of *Rasa Taragini.*

Preparation of *Rasakarpura Drava* with alcohol (Hydro-Alcoholic) media by following procedure of *RasaTaragini.*

To study physico-chemical properties and antimicrobial activity of *Rasakarpura drava.*

Modern Review – Mercuric Chloride

Rasakarpura is a mercurial salt, with chlorine mercury forms mercuric chloride ($HgCl_2$) Mercuric chloride (formerly *corrosive sublimate*), is the chemical compound with the formula $HgCl_2$. This white crystalline solid is a laboratory reagent. It is not used as commonly as once was the case because, due to its solubility in water, it is highly toxic. It is a molecular compound.

MATERIALS AND METHODS:

Materials include:-

- Collection of Major raw drugs
- Collection of Associated raw drugs.
- Equipments required for procedures.

1. Major raw drugs:-

Parada (Mercury), conc. Sulphuric Acid, Citric Acid, *Saindhav Lavana*(Rock Salt) are the major drugs collected from Authenticated pharmacy according to *Grahyaagrahyatva* mentioned in *Rasa* texts and authenticated by their experts.

2. Associated raw drugs:-

Rasona Kalka (Paste of Allium sativum), *Shudha Churna* and *Saindhava lavana* were the drugs used for *Samanya shodhana* of *Parad* and was collected from local market.

3. Equipments:-

Khalwa Yantra – It was used for *Samanya Shodhan* of *Parada* and to prepare *Kajjali.*

Valuka Yantra - It provide uniform Heat to *kupi*. Iron made *Valukayantra* was used for these preparations.

Sand - The sand particle size must be moderate and similar in size.

Kach Kupi - For *paka kriya* brown coloured beer bottle was used, total capacity of bottle was 650ml.

Gas burner , Glass flask, Conical flask - For preparation of *Parada Churna*.

Pyrometer – For measuring the temperature at different stage of *Kupipakva*.

Trays & Spatula – To collect and to scrap the medicine during various steps of the *Kupipakva* process.

Brick–Cork - This brick-cork was prepared by rubbing the brick piece against rough surface. It must be narrower at the end and round shaped at the base. Its shape should resemble the cone. It is useful for the *mudrana* procedure.

Knife - After removing the *kach-kupi* from the *Valuka yantra*, knife was required to remove the closely adherent layers of clay and cloth.

Glass rod -Glass-rod was required for gently striking the bottle so that the layers of *pakva rasa* from the bottle.

Cloth -*Manjarpath* cloth was used for *kapadmitti* of bottles.

Clay - Plain "*Multani-Mitti*" with water was used. It was sticky enough for the wrapping process.

Match box –For ignition of fire.

Torch - To see inside the *kupi* for different stages of *Kajjali.*

Kerosene - Kerosene soaked cloth piece was used every time while breaking the bottle.

Methods (Procedure) includes:-

The whole method of preparation includes:

- *Shodhana of Parada*.
- Preparation of *Parada churna*.

- Preparation of *Rasakarpura* by Kupipakva Method.
- Preparation of *Bhutaghna Chakrika*.
- Preparation of *Rasakarpura Drava* by Method 1 (water media).
- Preparation of *Rasakarpura Drava* by Method 2 (alcohol media).

1. Shodhan of Parada[1]:

- *Parada shodhan* was done with *Rasona, Katika Churna and Saindhav Lavana* in *Khalva Yantra* for one month.

2. Preparation of Parada Churna by Nirjalikarana Method[2] :

It is a *Parada Churna* preparation contains,

- *Shuddha Parada* (1 part) and Conc. Sulphuric Acid (1.5 part).
- This was Mix together in Glass Beaker and given heat, stirred mixture with Glass rod till *Parada Churna* obtained.

3. Preparation of *Kajjali* for *Kupipakva* Method :

It is a Nirgandh Niragni Khalvi Rasayana preparation contains,

- *Parada Churna* and *Saindhava Lavana.*
- This was triturated in Porcelain *Khalwa Yantra* till *Siddhi Lakshanas* were obtained.

4. Preparation of *Rasakarpura* by Kupipakva Method[2] :

- *Kajjali* for *Kupipakva* method was taken and then filled it into a *Kacha Kupi*, kept in *Valuka yantra* and heated over agni, for 3 *Prahar by Kramagni Tapa* but *tivragni* not used. The product was collected by breaking of *Kupi* after self cooling.

5. Preparation of Bhutaghna Chakrika[3] :

- 73 parts of Rasakarpura and 38 parts of *Nimbukamla* are taken in a *Kalwa Yantra* and mixed together.
- After proper mixing *Vati's* of 6 *ratti* size (750 mg) are prepared and stored.

6. Preparation of *Rasakarpura* Drava by 1st Method[4] :

- A Conical flask was taken; it was filled with 1.2lts of distilled water.
- To this 1 Bhutaghna Chakrika(6 ratti-750mg) was added.
- This mixture was properly stirred using a glass rod for about 15 mins and then used.

7. Preparation of *Rasakarpura* Drava by 2nd Method[5] :

- 4gms of *Rasakarpura* was taken in a glass rod and added to a conical flask containing 30 ml of ethyl alcohol.
- It was mixed well for about 10-15 mins.
- 3.2ml of this solution was pippetted out and added to another conical flask containing 1.2lts of distilled water.
- This was stirred well using a glass rod for about 5 mins and *Rasakarpura* Drava was formed.

PREPARATION OF *BHUTGHNA CHAKRIKA*

Reference : *Rasatarangini* 6 /87-89

Materials : *Rasakarpura* 73 parts (5.110 gm), *Nimbukamla* (citric acid) 38 parts (2.660 gm)

Equipments: Porcelain mortar and pestle, weighing balance, glass rod.

Procedure:

- 73 parts of *Rasakarpura*(5.110 gm) and 38 parts of *Nimbukamla* (citric acid) (2.660 gm) are taken in a porcelain *Kalwa Yantra* and mixed together.
- After proper mixing *Vati's* of *6 ratti* size (750 mg) are prepared and stored.

Precautions:

- Metal containers were not used throught the process as *Rasakarpura* reacts with it.
- Mask and gloves were used for protection

PREPARATION OF *RAKAKARPURA DRAVA*

Rasakarpura Drava can be prepared in 2 ways i.e in Aqueous media and in aqua-alcohol media.

PREPARATION OF *RASAKARPURA* DRAVA USING WATER (METHOD I)

भूतन्धचक्रिकामेकां शततोलकसंमिते ।
जले क्षिपेद् विद्रुतायां द्रवं तु विनियोजयेत् ॥ ९६॥
रसकर्पूरमानात्तु व्दिसहस्त्रगुणाभ्भसा ।
द्रवोऽयमेवं निर्दिष्टो यथा योगं प्रयोजयेत् ॥ ९७॥ –
रसतरंगिणी ६/ ९६–९७.

Materials : 1(750 mg) *Bhutaghna Chakrika*, 100 *Tolas* of Water (1.2 lts)
Method : Dissolution
Equipments : Conical flask, glass rod

Procedure:
- A Conical flask was taken; it was filled with 1.2lts of distilled water.
- To this 1 *Bhutaghna Chakrika* (6 *ratti*-750mg) is added.
- This mixture was properly stirred using a glass rod for about 15 mins and then used.

Precautions:
- Gloves were used during the process
- Stirring was done carefully

PREPARATION OF *RASAKARPURA* DRAVA IN AQUA-ALCOHOL MEDIA.

द्वात्रिंशदगज्जकमित रसं कर्परसंज्ञकम् ।
सार्धद्वितोलकमिते सारे कोहलपूर्वके ॥९८॥
द्रावयेद्रसतन्त्रज्ञः कुप्यामथ निधापयेत्।
द्रवस्यास्य समादाय त्वष्टमांशमितं द्रवम्॥९९॥
मेलयेद्रत्सलिले शुध्दे शततोलकसंमिते ।
द्रवोऽयमपि संवृत्तो द्विसहस्त्रणाभ्भसा ॥ १००॥
–रसतरंगिणी ६/ ९८–१००.

Materials :*Rasakarpura* - 32 gunja (4 gms), Alcohol - 8 mash (30 ml), Water of 100 Tolas (1.2 lts)
Method : Dissolution
Equipments : 2 conical flasks, Pipette, glass rod.

Procedure:
- 4gms of *Rasakarpura* was taken in a glass rod and added to a conical flask containing 30 ml of ethyl alcohol.
- It was mixed well for about 10-15 mins.
- 3.2ml of this solution was pippetted out and added to another conical flask containing 1.2lts of distilled water.
- This was stirred well using a glass rod for about 15 mins and *Rasakarpura* Drava was formed.

Precaution:
- The *Rasakarpura* was taken in a glass rod,spatula made out of steel etc were not used as *Rasakarpura* reacts with it.
- Pipetting was done using a rubber cork , mouth pipetting should not be done as the Solution is toxic.
- Mask and Gloves were worn throught the process.
- Stirring was done carefully.

OBSERVATIONS AND RESULTS:

BHUTGHNA CHAKRIKA:
Observation:
- While mixing *Rasakarpura* and Nimbukamla(citric acid) no reaction takes place.
- Vati can be easily prepared.

Properties of Bhutaghna Chakrika:
Color : White
Consistency : Hard
Form : Pellet
Odour : faint odour of citric acid

***Rakakarpura Drava* in Water Media(METHOD 1):**
Observation:
The Bhutaghna Chakrika did not dissolve easily and hence stirring was necessary.

Properties of *Rasakarpura* Drava Using Water:
Color - Colorless
Form - Liquid
Odour - Odourless

***Rasakarpura Drava* in Aqua-Alcohol Media (METHOD 1):**
Observation:
- When *Rasakarpura* was mixed in the alcohol(Kohala) the solution turned turbid and once 1.2 lts of water was added to it the solution became colourless.
- The *Rasakarpura* did not mix easily and stirring was required.

Properties of *Rasakarpura* Drava Using Kohala(Alcohol)**:**
Color - Colorless
Form - Liquid
Odour - odourless

PHYSICO CHEMICAL ANALYSIS OF *RASAKARPURAA*:

Sr. No.	Name of Test	Result
1	Colour	White
2	Form	Crystalline
3	Odour	Mildly irritant
4	Consistency	Hard
5	PH	4.69
6	Loss on drying	8.57
7	Solubility in water	99.56%
8	Solubility in Alcohol	98.97%
9	Total ASH	0.23%
10	Acid insoluble	‹0.01%

Table showing result of XRF-Ray-Fluorescence (XRF):-

Sr.No.	Element	Mass(%)	2sigma(%)
1	Mercury	73.535	0.066
2	Chlorine	25.990	0.023
3	Oxygen	0.073	0.014
4	Strontium	0.402	0.076

BY METHOD 1:

A)Organoleptic Test :

Color - Colorless
Form - Liquid
Odour - Odourless

B)Refractive Index: 1.3324
C)Specific Gravity: 1.0005

BY METHOD 2:

A)Organoleptic Test :

Color - Colorless
Form - Liquid
Odour - Odourless

B)Refractive Index: 1.3328
C)Specific Gravity: 0.9999

Graph Showing peaks of X-ray diffraction.

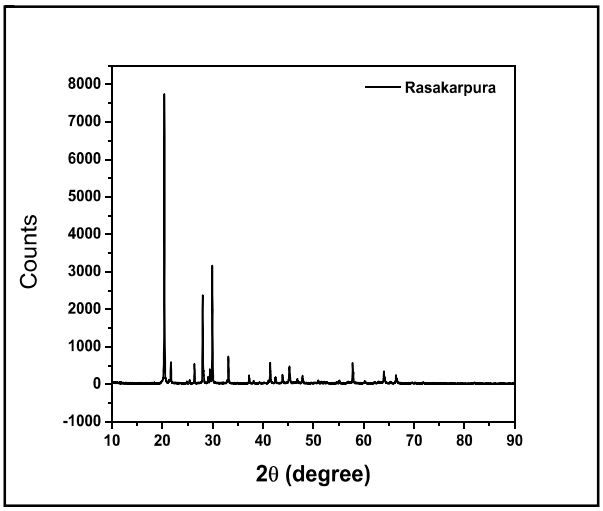


Table Showing results of X-ray diffraction.

Sample	Major Phase
Rasakarpura	$HgCl_2$

ANTIMICROBIAL STUDY:

A) Antimicrobial study for Sample A:-

Sr.No	Bacteria/Fungi	Diameter of inhibition zone in (mm)
1	Staphylococcus aureus	19
2	Escherichia coli	23
3	Candida Albicans	10

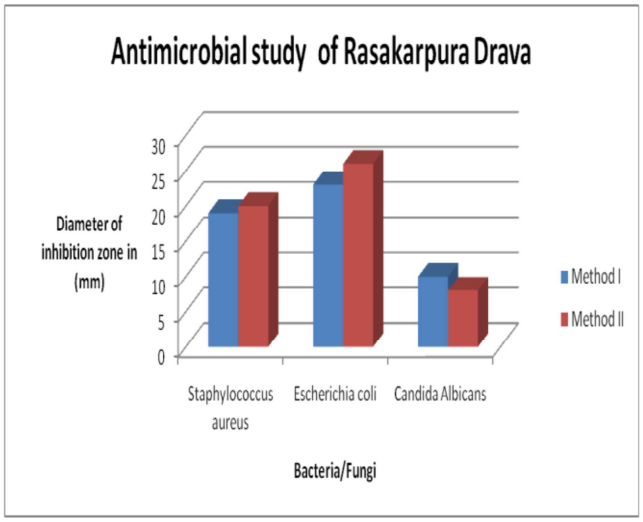
B) Antimicrobial study for Sample B:-

Sr.No	Bacteria/Fungi	Diameter of inhibition zone in (mm)
1	*Staphylococcus aureus*	20
2	*Escherichia coli*	26
3	*Candida Albicans*	08

Sample A and Sample B has shown significant antimicrobial activity.

1) Sample A shown 19mm of zone of inhibition against *Staphylococcus aureus*

2) Sample B shown 20mm of zone of inhibition against *Staphylococcus aureus*
3) Sample A shown 23mm of zone of inhibition against *Escherichia coli*
4) Sample B shown 26mm of zone of inhibition against *Escherichia coli*
5) Sample A shown 10mm of zone of inhibition against Candida Albicans
6) Sample B shown 08mm of zone of inhibition against *Candida Albicans*
6) Here, noted that as compaired to Sample B, Sample A had shown less zone of inhibition against *Staphylococcus aureus.*
7) Here, noted that as compaired to Sample B, Sample A had shown less zone of inhibition against *Escherichia coli.*
8) Here, noted that as compaired to Sample A, Sample B had shown less zone of inhibition against *Candida Albicans.*



DISCUSSION:

Rasakarpura drava can be used as tropical application.

Raskarpura drava was prepared by two methods changing the liquid medium.

- Raskarpura drava in Aqua. Method 1
- Raskarpura drava in Aqua-alcohol. Method 2

For both the types of *Rasakarpura* drava, the method used was simple dissolution.

In the case of aqueous solution, *Bhutaghna chakrika* was dissolved in a specified quantity (100 toles) of distilled water by a process of stirring with a glass rod. Obtained product was colourless, odourless liquid.

In the case of alcoholic solution, instead of Bhutaghna chakrika *Rasakarpura* was directly dissolved in the specific amount of alcohol as mentioned in *Rasatarangini*. This alcoholic solution was further diluted with water to get an aqua-alcoholic solution. It is in this diluted form that *Rasakarpura drava* is tropically used. This *Rasakarpura drava* was turbid with an alcoholic odour, it became colourless and the odour diminished when diluted.

In total it was found that the test drug was possesses excellent result against the organisms selected for the study. Chemically *Rasakarpura* is found to be $HgCl_2$ as a major phase. Various studies carried out indicated that the solution of $HgCl_2$ is very effective as an antimicrobial agent.

In the preparation of *Rasakarpura* Drava by method 1(sampleA), *Bhutaghna chakrika* is important. In *Bhutaghna chakrika Nimbukamla* (Citric acid) is an additional ingredient, it also possess weak anti-microbial activity. It was found that a lower pH enhances the antimicrobial activity of organic acid like citric acid. It is possible that citric acid when added with *Rasakarpura* which is highly acidic to form Bhutaghna chakrika may have an improved anti-microbial activity and it is use for preparation of sample 1.

In the preparation of *Rasakarpura Drava* by method 2(sampleB), Alcohol is important. Alcohol itself is also having some anti-microbial property especially anti bacterial and anti fungal effect.

CONCLUSION

In the present research work on pharmaceutical, analytical and antimicrobial study it could be concluded that:

Analytically both *Rasakarpura* Drava showed almost near about simmilar result.

Anti-microbial evaluation of *Rasakarpura* drava, prepared by two methods samples proved that both samples were highly effective. However *Rasakarpura* drava prepared by method 2 i.e. dissolving *Rasakarpura* in alcohol and then water was found to be most effective. The products were useful against *S. aureus, C. albican and E. coli.*

These observations have opened a new path for the development of user friendly tropical application of *Rasakarpura Drava* in various kinds of skin infections like psoriasis, Eczema, Bedsores etc. and day to day life product like hand wash,

instrument sterilizer etc. A detailed investigation on dermatological toxicity of the product is essential but considering the high potential of *Rasakarpura Drav*a saftey may not be a major issue.

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