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PHARMACEUTICO- ANALYTICAL STUDY OF RAKTA PRAVARTHANA CHURNA

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ABSTRACT

Rakta Pravarthana Churna is an Ayurvedic formulation designed for external application during *Rakta Mokshana* to address *apravarthana* or clotting tendencies. The research aims to evaluate the pharmaceutical and analytical aspects of this formulation, considering its potential applications in coagulation disorders from a broader perspective. **Aims & Objectives**: The primary aim of this study is to conduct pharmaceutical and analytical studies on *Rakta Pravarthana churna*, a compound formulation for regularising blood flow. **Materials & Methods**: The pharmaceutical study involves preparation of churnas of all the herbal ingredients, procuring & processing of *Gruhadhuma*, and finally the compound *churna* preparation. The analytical study comprises organoleptic, physicochemical, and chromatographic evaluations. **Results**: The preparation of *Rakta Pravarthana Churna* involved continuous trituration, resulting in a greenish-black powder with a characteristic smell of *Karpura*, and possessing *katu-kashaya rasa*. The quantity obtained was 750g. Analytical parameters and HPTLC provided relatable results. **Discussion:** The pharmaceutical study notes complexities such as varying hardness of individual drugs, the usage of *Gruhadhuma*, its sourcing, safety

and appropriateness, and other challenges in size reduction. Organoleptic evaluation reveals the formulation's unique features, including its katu rasa, supporting its role in regulating blood flow. HPTLC analysis demonstrates distinct compound patterns. **Conclusion**: In conclusion, this study offers the standardization of *Rakta Pravarthana Churna* & provides a foundation for further research in the field of Ayurvedic preparations pertaining to blood flow regulation.

KEYWORDS: *Rakta pravarthana*, coagulation, *Gruhadhuma*, pharmaceutical study, analytical parameters, standardization.

INTRODUCTION

Coagulation disorders encompass a broad spectrum of conditions characterized by abnormalities in the clotting mechanism of blood. Any disruption in this delicate equilibrium can lead to coagulopathies which have significant clinical implications like DVT, pulmonary embolism and thrombophilia. In these conditions, the coagulation cascade becomes dysregulated, resulting in an excessive formation of clots that can block blood vessels and impair blood flow to vital organs.

Ayurveda, the ancient Indian Life science and system of medicine has elaborate discussions on *Rakta dhatu*, its functions in normalcy, signs and symptoms on vitiation, its disorders, their treatment strategies, and efficient therapeutic formulations. In this context, *Rakta mokshana* is stated to be the prime modality of treatment for *raktaja vyadhis*[1], and to ensure that *raktamokshana* itself is not hindered by preexisting clotting tendencies due to *rakta dushti*, a set of herbo-mineral drugs for *Rakta pravarthana* activity has been mentioned by Acharya Sushrutha.[2] This formulation is prepared in the form of *churna* and intended for external application on the site of *raktamokshana*. Considering a broader perspective, this formulation may find further applications in coagulation disorders, and as an initial step its pharmaceutical & analytical evaluation is undertaken in the present study.

AIMS AND OBJECTIVES

Rakta pravarthana churna, a polyherbo-mineral formulation with potential modulatory action on the blood coagulation mechanism is taken up for the present study. This work includes pharmaceutical and analytical studies on *Rakta Pravarthana churna*.

MATERIALS AND METHODS

PHARMACEUTICAL STUDY

- Pharmaceutical source: Raw drugs required for the preparation of *Rakta Pravarthana Churna* were collected from SDM Pharmacy, Udupi and authentication is done by the subject experts at Sri Dharmasthala Manjunatheshwara College of Ayurveda, Udupi.
- The preparation of *Rakta Pravarthana Churna* was carried out in practical hall of dept. of Rasa Shastra & Bhaishajya Kalpana, SDMCA, Udupi.
- *Rakta Pravarthana Churna*, a formulation from *Sushruta Samhita* was prepared according to general principles of *Sharngadhara Samhita*.
- Preparation of *Rakta Pravarthana Churna* includes

1. Preparation of *churnas* of *Ela, Karpura, Kushta, Tagara, Paata, Bhadradaaru, Vidanga, Chitraka, Shunti, Maricha, Pippali, Haridra, Arka, Naktamala.*
2. Preparation of *Aagaradhuma Churna*
3. Preparation of *Rakta Pravarthana Churna*

Table 1: Ingredients of *Rakta Pravarthana Churna.*

Drug Name	Botanical Name/ Source	Family Name	Part used	Ratio
Ela	*Elettaria cardamomum Maton*	Scitamineae	Seeds	1 part
Karpura	*Cinnamomum camphora Nees*	Lauraceae	Extract	1 part
Kushta	*Saussurea lappa*	Asteraceae	Root	1 part
Tagara	*Valeriana wallichii*	Valerianaceae	Root	1 part
Paata	*Cissampelos pareira*	Menispermaceae	Root, stem	1 part
Bhadradaaru	*Cedrus deodara Roxb*	Pinaceae	Bark	1 part
Vidanga	*Embelia ribes Burm*	Myrsinaceae	Fruits	1 part
Chitraka	*Plumbago zeylanica Linn.*	Plumbaginaceae	Root	1 part
Shunti	*Zingiber officinale*	Scitamineae	Rhizome	1 part
Maricha	*Piper nigrum Linn.*	Piperaceae	Fruit	1 part
Pippali	*Piper longum Linn.*	Piperaceae	Fruit	1 part
Aagaradhooma	*house soot*	Not applicable		1 part
Haridra	*Curcuma longa Linn.*	Scitamineae	Rhizome	1 part
Arka	*Calotropis gigantea Linn.*	Asclepiadaceae	Flower bud	1 part
Naktamala	*Pongamia pinnata*	Fabaceae	Fruit	1 part

Step 1: Preparation of Churnas of *Ela, Karpura, Kushta, Tagara, Paata, Bhadradaaru, Vidanga, Chitraka, Shunti, Maricha, Pippali, Haridra, Arka, Naktamala*

Reference: *Sharangadhara Samhita*[3]

Equipment: *Tulayantra, ulukhala yantra*, grinder, sieve, vessels

Principle involved: Size reduction Size separation

Method of preparation: The drugs were collected in required quantities and dried completely. Then they were taken separately and pounded manually in *ulukhala yantra* and further made into fine powder using mixer grinder and filtered through sieve to obtain uniform fine powder. The churnas were weighed and stored in separate labelled airtight containers.

Step 2: Preparation of *Aagaradhuma/grihadhuma churna*

Equipment: *Loha patra, tulayantra, ulukhala yantra*, sieve, vessels

Ingredients: Carbon soot

Principle involved: Combustion, size reduction and size separation

Method of preparation: Charcoal soot was collected from the furnace of SDM pharmacy. It was dried and then heated in a *loha patra* to remove any remaining moisture and ensure complete soot formation. Then it was powdered using *ulukhala*, filtered through sieve and stored in airtight container.

Observation: The collected soot and charcoal consist of incompletely burnt irregular shaped pieces. On heating the charcoal pieces in *loha patra* they become very brittle and powdery. After proper pounding and sieving, very fine black powder of soot is obtained which is light and soft in nature.

Step 3: Preparation of *Rakta Pravarthana Churna*

Reference: Sushruta Samhita[2]

Equipments: *Tula yantra, khalva yantra*, vessel, spoon, plastic containers

Ingredients: Equal quantity of each of Ela churna, Karpura churna, *Kushta churna, Tagara churna, Paata churna, Bhadradaaru churna, Vidanga churna, Chitraka churna, Shunti churna, Maricha churna, Pippali churna, Aagaradhuma churna, Haridra churna, Arka churna, Naktamala churna.*

Principle involved: Mixing and homogenisation.

Method of preparation: Equal quantity of all the churnas were taken in *khalva yantra* one by one and *mardana* was done to mix the ingredients. It was again mixed well using spoon. The prepared *churna* was weighed and stored in airtight plastic containers.

Precaution: The *churna* should be in homogenous form, hence continuous trituration and mixing is necessary. While adding and mixing the individual powders, it must be ensured that all the equipment used are free from any dust and moisture. The final *churna* is immediately stored in airtight plastic containers.

ANALYTICAL STUDY: The analytical study is pivotal in ensuring quality, safety, and efficacy of the formulation. Organoleptic, physicochemical, and chromatographic evaluation of *Rakta Pravarthana Churna* was performed. Physicochemical analysis includes ash value, acid insoluble ash, water soluble ash, water soluble and alcohol soluble extractive values and loss on drying. Chromatography technique employed was HPTLC.

RESULTS

Observations: Continuous thorough trituration was required for making a homogenous mixture. The colour and smell of the mixture underwent a change with the addition of each component *churna.* At the end, *Rakta pravarthana churna* was a greenish-black coloured fine powder. It had a characteristic smell predominantly of that of *Karpura.*

Table 2: Results of *Rakta Pravarthana Churna* preparation.

Drug	**Quantity**
Ela, Karpura, Kushta, Tagara, Paata, Bhadradaaru, Vidanga, Chitraka, Shunti, Maricha, Pippali, Aagaradhuma, Haridra, Arka, Naktamala	50g of each drug taken
Rakta Pravarthana Churna	Quantity obtained **750g**

Table 3: Organoleptic characteristics.

Organoleptic characteristics	***Rakta Pravarthana Churna***
Colour	Greenish black
Taste	*Katu, Kashaya*
Smell	Characteristic, predominantly of Karpura
Consistency	Fine powder

Table 4: Results of analytical parameters.

Parameter	**Results (Avg±SD)**
Loss on drying	15.71±0.02
Total Ash	5.67±0.0
Acid Insoluble Ash	0.68±0.02
Water soluble Ash	2.66±0.03
Alcohol soluble extractive value	11.16±0.01
Water soluble extractive value	16.28±0.23

Chromatography -HPTLC

Table 5: R_f values of Ethanol extract of Rakta Pravarthana Churna.

Short UV	Long UV	White light	Post derivatisation
-	0.08 (F.green)	-	-
0.15 (Green)	-	0.15 (Yellow)	-
-	0.17 (F.blue)	-	0.17 (Red)
-	0.22 (F.blue)	-	-
0.26 (Green)	0.26 (F.blue)	-	-
-	0.30 (F.blue)	-	-
0.34 (Green)	-	-	-
-	0.40 (F.blue)	-	-
0.46 (Green)	0.47 (F.blue)	-	-
0.52 (Green)	-	-	-
-	0.54 (F.blue)	-	-
-	-	-	-
0.66 (Green)	-	-	0.66 (Purple)
0.71 (Green)	-	-	-
0.80 (Green)	-	-	-
0.83 (Green)	-	-	-
-	-	-	0.85 (blue)

*F – Fluorescent; L –Light; D – Dark

Herbal drugs of *Rakta Pravarthana Churna*

Fig. 1: *Ela* raw drug

Fig. 2: *Karpura* raw drug

Fig. 3: *Kushta* raw drug

Fig. 4: *Tagara* raw drug

Fig. 5: *Paata* raw drug

Fig. 6: *Bhadradaaru* raw drug

Fig 7: *Vidanga* raw drug

Fig 8: *Chitraka* raw drug

Fig 9: *Shunti* raw drug

Fig 10: *Maricha* raw drug

Fig. 11: *Pippali* raw drug.

Fig. 12: *Haridra* raw drug.

<table>
<tr><td>  </td><td>  </td></tr>
<tr><td>Fig. 13: Arka raw drug.</td><td>Fig. 14: Naktamala raw drug.</td></tr>
<tr><td colspan="2">Preparation of Aagaradhuma Churna</td></tr>
<tr><td>  </td><td>  </td></tr>
<tr><td>Fig. 15: Collected gruhadhuma</td><td>Fig 16: Pounding Gruhadhuma in khalvayantra.</td></tr>
<tr><td>  </td><td>  </td></tr>
<tr><td>Fig. 17: Heating it for complete soot formation.</td><td>Fig. 18: Gruhadhuma Churna.</td></tr>
</table>

Preparation of *Rakta Pravarthana Churna*

Fig. 19: Individual ingredients.

Fig. 20: Stirring in vessel

Fig. 21: Weighing the Compounded *churna.*

Fig. 22: Packing and storing in plastic containers/covers.

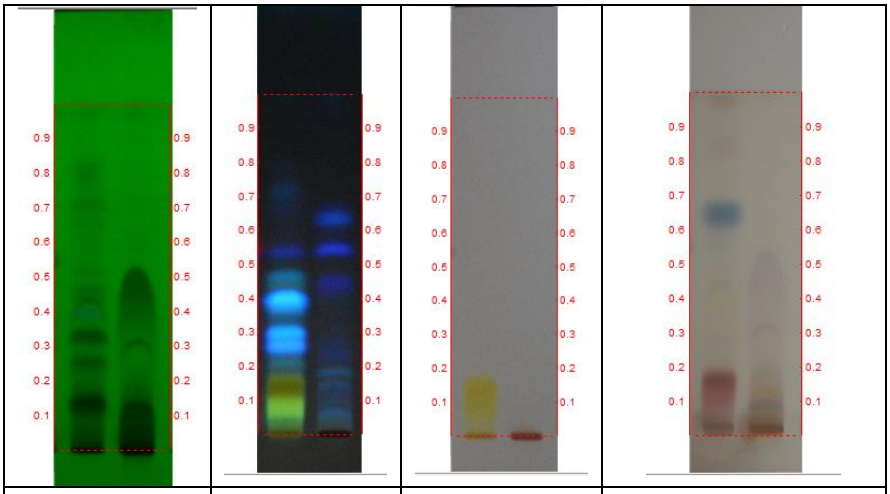

Fig. 23: HPTLC photo documentation of ethanol extract of Rakta Pravarthana Churna.

Ethanol extract of *Rakta pravartana churna* – 5µl

Solvent system – Toluene: Ethyl acetate (9.0: 1.0)

DISCUSSION

Rakta Pravarthana Churna is formulated based on the reference of the treatment for *apravrutti* of *rakta* in Sushruta Samhita. The c*hurna* was prepared according to the general method of preparation. The hardness and nature of the individual drugs varied and was perceivable during pounding of drugs. For example: drugs like *Ela, Vidanga, Pippali* required simple pounding whereas fibrous drugs like *Paata, Bhadradaru* needed more crushing and grinding. Very hard and fibrous drugs like *Tagara, Kushta, Chitraka* gave lesser yield of churna as size reduction for the entire drug was difficult to achieve. The odour and other characteristics of individual drugs were observed during the procedure. *Karpura* needs to be stored in a completely sealed container and preferably stored in a cool dark place as it is a highly volatile drug and tends to sublime even at room temperatures. Size reduction up to fine powder form was achieved for all herbal drugs. Particle size reduction increases the surface area of drug exposed thereby increasing the pharmacological activity of the drug.

Aagaradhuma/ Grihadhuma: Classically, *grihadhuma*, as its name implies, refers to the soot collected from the walls and chimneys of homes. However, collecting *grihadhuma* from such sources has become exceedingly challenging in contemporary times. For this study, charcoal soot was collected from the furnace of SDM pharmacy. As the collected soot contained pieces of incompletely burnt charcoal, it was initially dried and then subjected to heating in a *loha patra* to eliminate any remaining moisture and ensure complete formation of soot. The

process rendered the charcoal material extremely brittle and powdery. Subsequently, the material underwent careful pounding and sieving, resulting in a fine, jet-black coloured powder.

As per Acharya Sharangadhara's description of *anuktadravya pramana*, if the specific quantity or proportion of drugs is not specified in any compound formulation, then equal quantity of each ingredient has to be taken. Here the fine powder of each ingredient is separately prepared and then taken in equal quantity for the preparation of *Rakta Pravarthana Churna.*

Continuous mixing is required to achieve a homogenous mixture of the churna without any lumps. The colour and smell of the mixture underwent a change with the addition of each ingredient. Finally, *Rakta pravarthana churna* obtained was a greenish-black coloured fine powder with a characteristic smell predominantly of that of *Karpura.*

The evaluation of the organoleptic characteristics of *Rakta Pravarthana Churna* brings to light the inherent uniqueness and therapeutic intentions of the formulation. *Rakta Pravarthana Churna* exhibited a greenish-black hue. The colour serves as an indicator of the different herbal and mineral constituents. *Rakta Pravarthana Churna's* taste is characterized by *katu* and *kashaya rasa* (pungent and astringent), aligning precisely with its intended purpose. The predominance of *katu rasa*, ascribed with the action of *shonita sanghata bhinatti*, possibly translating to clot lysis, lends credence to its role in facilitating *rakta pravarthana*. The churna emanates a characteristic aroma primarily derived from *Karpura.*

Rakta Pravarthana Churna exhibited a loss on drying of 15.71%, indicating a relatively higher moisture content. This might be attributed to the presence of more moisture-absorbing ingredients. The total ash content measured 5.67% which signifies minimum concentration of mineral components. Analysing the acid insoluble ash and water-soluble ash, it exhibited values of 0.68% and 2.66% respectively. It had an alcohol soluble extractive value of 11.16% and water-soluble extractive value of 16.28%. This divergence might indicate *Rakta Pravarthana Churna's* ability to release more water-soluble bioactive compounds.

High-Performance Thin Layer Chromatography (HPTLC)

In the HPTLC report, the Rf (Retention Factor) values of the sample from the ethanol extract of *Rakta Pravarthana Churna* was measured under different conditions of UV light and post-

derivatization. The values obtained for the samples under different conditions reveal distinct chromatographic patterns.

Rf value at short UV revealed the presence of 9 spots at the Rf values of 0.15, 0.26, 0.34, 0.46, 0.52, 0.66, 0.71, 0.80 and 0.83. **Rf value at long UV** revealed the presence of 8 spots at the Rf values of 0.08, 0.17, 0.22, 0.26, 0.30, 0.40, 0.47 and 0.54. **Rf value at white light** revealed the presence of one spots at the Rf values of 0.15(yellow). **Post derivatization** revealed the presence of 3 spots at the Rf values of 0.17, 0.66 and 0.85. **Densitometric scan at 254nm** shows maximum area at Rf value 0.16 i.e., 24.17 %. **Densitometric scan at 366nm** shows maximum area at Rf value 0.47 i.e., 36.46 %. **Densitometric scan at 620nm** shows maximum area at Rf value 0.75 i.e., 46.10 %. The observed Rf values suggest the presence of distinct compounds with varying affinities for the stationary phase (silica gel) and the mobile phase (ethyl acetate-methanol). The presence of fluorescent compounds suggests the possibility of aromatic or conjugated systems in the churna. The variations in Rf values under different UV and light conditions highlight the presence of compounds with different polarities and molecular structures. The Rf values might correspond to the presence of active compounds with specific therapeutic activities.

CONCLUSION

From pharmaceutical study it can be concluded that the transformation of raw ingredients into fine powders involved the application of various principles of size reduction and size separation. The formulation under study contains *Gruhadhuma* and, in this context sourcing of soot for medicinal formulations holds substantial significance. Utilizing soot raises considerations regarding its procurement, safety, and appropriateness for the intended usage. This itself forms a topic for further research.

Analytical investigations provide valuable insights into the composition, properties, and interactions of the individual components. HPTLC added another layer of understanding, revealing the presence of diverse compounds with varying affinities. Thus, the analytical study contributes to the standardization and quality control of the formulation and serves to enhance our understanding of its underlying pharmacological effects.

In conclusion, the pharmaceutical & analytical study of *Rakta Pravarthana Churna* provides a base for further research and subsequently aids in adopting such formulations into Ayurvedic practise enriching both the science and healthcare.

Conflicts of interest: There are no conflicts of interest.

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