

DOI-10.53571/NJESR.2022.4.5.10-19

## Analytical Standardization for “*Rasa Chenduram*” Through Characterization by Physico-Chemical Analysis and Fourier Transform Infrared Radiation(FTIR) Analysis.

K. Balagurusamy\*, S.Semalatha\*\*, S.Balamurugan\*\*\*

\*Principal, \*\* Lecturer, \*\*\*Lecturer,

Velumailu Siddha Medical College and Hospital Sriperumbudur

[dr.k.balagurusamy@gmail.com]

(Received:2April2022/Revised:15 April 2022/Accepted:29April2022/ Published: 2May2022)

### Abstract

Siddha System of medicine is one of the oldest medicinal therapy that brings out the most effective treatment for various diseases. According to this Indian System of medicine, *Chenduram* is a red coloured powder composed of herbal, metal and mineral products. This paper deals with the standardization of the Siddha drug “*RASA CHENDURAM*”. It comprises of ingredients such as *Rasam* (hydragyrum - Mercuric quicksilver), *Gandhagam* – Sulphur and *Thalagam* (yellow arsenic trisulphide). *Rasa chenduram* has been used for the treatment of various ailments like Cancer, Leprosy, and Syphilis. The major bottleneck in the wider acceptance of Siddha drugs from developing countries is due to the lack of standardization of the drug. The main aim of this study is to standardize the Siddha drug “*Rasa Chenduram*” through physicochemical analysis and Fourier Transform Infrared Radiation (FTIR) analysis. The result obtained from the physicochemical evaluation reveals that the specific gravity of RC was found 0.967 pH value 4.79% , total ash value of RC was found 1.8%. In which the water-soluble ash was less than 1%, acid-soluble ash less than 1%, water-soluble extractive 2.3% and alcohol soluble extractive 1.91%. Similarly, loss of drying value at 52.02%. pH is an important indicator of solvent solubility and the ability to cross the biological barrier. From the result of the study, it was evident that the Siddha formulation RC complies with standards and may be used for Cancer, leprosy, syphilis etc. This analysis and quality control of Herbo-mineral formulation are moving toward the integrative and comprehensive direction, to better address the inherent holistic nature of Herbo-mineral medicine.

**Keywords:** Siddha Medicine, *Rasa Chenduram*, FTIR, Physio chemical analysis.

## 1. Introduction

The Siddha system is a treasure house of a secret science embodying several medicines to cure several diseases. This Siddha system of medicines is being utilized by Indians and has also gained attention worldwide due to its long-term benefits in terms of overall wellness with no side effects<sup>[1]</sup>. Medicines like *Parpam*, *Chenduram*, *Kattu* and *Padhangam* are like “lifesaving” and “miracle”. These were prepared by the siddhars on the basis of Nano Medicine. Recent advances in science explored that the nano particles find the potential usage in bio-medical field, especially in cancer and many degenerative diseases<sup>[2]</sup>. In the present investigation, validation of a Siddha herbo-mineral preparation *Rasa Chenduram* was done, which indicated Cancer, Leprosy, Syphilis, by analyzing the Physio-chemical properties and Fourier Transform Infrared Spectroscopy (FTIR) analysis. Though this medicine has been traditionally used, there is no scientific validation reported for this formulation. The main aim of the present study is to systematically standardize the drug *Rasa Chenduram* (RC) using physiochemical and FTIR techniques, in order to generate the evidence-based data about this novel formulation so as to provide valuable information to the researchers, who will be working on this formulation.

## 2. Materials and methods

### Ingredients:

The ingredients of “*Rasa Chenduram*” are 1. *Rasam* (Mercury) 2. *Ghanthagam* (Sulphur), 3. *Thaalagam* (Arsenic trisulfide) were purified 4. Egg white as mentioned in *pulipani vaithiyam 500, Page no 28*.

### 2.1 Drug collections:

All the raw materials were obtained from the country shop, Ramaswamy Chetty, Parry’s, Chennai.

### 2.2 Identification and authentication:

All the raw drugs were identified and authenticated by Gunapadam experts in Velumailu Siddha medical college, Sriperumbudur. The specimen sample of each raw drug has been kept in the Gunapadam department for future reference.

### 2.3 Purification of the drug

#### a) Rasam

Mercury is widely used in the Siddha system in several medicines in various forms. It is therapeutically used as an alternative, tonic, laxative, diuretic and in venereal diseases.

### b) Method of Rasam Purification

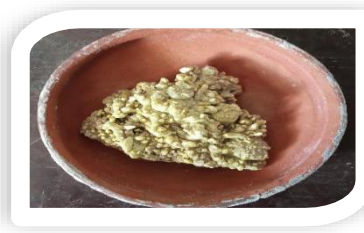
The raw material is ground well with brick powder till its colour turn's to white and is removed. Again ground with turmeric powder till its colour changes to black and the turmeric powder will remain separate and is removed. Finally, semi-purified *Rasam* is boiled with the *Acalyphaplant* juice. Washed, dried and used<sup>[3]</sup>.

### c) Sulphur:

Sulphur is widely used in the Siddha system in several medicines in various forms. It is therapeutically used as an alternative, tonic, for all types of skin diseases.

### d) Method of Sulphur purification

A sufficient quantity of butter was taken slightly heated and added 250 gm of Sulphur and melted at a temperature of 60-70C. The above-melted Sulphur is poured into an earthen container containing cow's milk. The Sulphur poured into the milk is allowed to cool. The above process is repeated thirty times, and fresh milk is used each time. The purified Sulphur after cooling is filtered and washed with the normal water<sup>[3]</sup>



### e) Thalagam

It is effective in the treatment of skin diseases, diseases of the head and tongue, fever with chills, Kapha diseases, urinary tract diseases, and venereal focus ulcers in the urethra.

### f) Method of purification:

1 Palam(35gm) of Thalagam was put in between the heap of lime stones and treated with the urine of a donkey ten times. take that thalagam wash in water and dry<sup>[6]</sup>.

### 2.4 Preparation of *Rasa Chendhooram* (RC)

*Rasam*(Mercury) and *Ghanthagam*(Sulphur), *Thaalagam* (Arsenic trisulfide) were purified separately as per Siddha literature. The purified metals were well-grounded. In a mud pot first transfer the contents of the egg and then purified metals mixture, now again transfer the contents of



another egg. Place the mud pot in the sand and ignite it using ten cow dungs. Collect the *chenduram* in mud pot and stored it in airtight container. The final product of *Rasa Chenduram* was obtained and labelled as RC. This preparation was used for further studies.

## **2.4 Organoleptic and physical properties**

The organoleptic characters of the sample drug were evaluated as per the standard procedure. 7.1 gm of the RC was taken and the colour, texture, particle size and other morphology were viewed by naked eye under sunlight. Then the result was noted.

## **2.5 physico-chemical analysis<sup>[4]</sup>:**

Physico-chemical investigations like pH value, Loss on drying at 105°C, solubility test, Determination of total ash, Determination of acid insoluble ash, Determination of water soluble ash, Determination of water soluble extractive, Determination of alcohol soluble extractive, have been done at The Tamilnadu Dr M.G.R Medical University, Anna salai, Guindy, as per the guide lines of WHO <sup>[5]</sup>.

### **2.6.1 pH value:**

Potential metrically, pH value is determined by a glass electrode and a suitable pH meter. The pH of the RC was written in the results column.

### **2.6.2 Loss on Drying:**

An accurately weighed 2 gram of *Rasa Chenduram* formulation was taken in a tarred glass bottle. The crude drug was heated 105<sup>0</sup> c for 6 hours in an oven till constant weight. The percentage moisture content of the sample was calculated with reference to the shade dried material.

### **2.6.2 Solubility test:**

A pinch of a sample [RC] was taken in a dry test tube and 2ml of the solvent was added and shaken well for about a minute and the result are observed. the test was done for solvents like distilled water, Ethanol, petroleum, ether, propylene glycol, Toluene, Benzene, chloroform, Ethyl alcohol, Xylene, and Carbon tetrachloride, and the result are observed individually.

### **2.6.4 Determination of acid insoluble ash:**

Ash above obtained was boiled 5min with 25ml of hydrochloric acid and filtered using an ash less filter paper. Insoluble matter retained on filter paper was washed with hot water and filter paper was burnt to a constant weight in a muffle furnace. The percentage of acid insoluble ash was calculated with reference to the air dried drug.

### 2.6.5 Determination of water soluble ash:

Total Ash 1gram was boiled for 5min with 25ml water and insoluble matter collected on an ash less filter paper was washed with water and ignited for 15 min at a temperature not exceeding 450<sup>0</sup>c in a muffle furnace. The amount of soluble ash is determined by drying the filtrate.

### 2.6.6 Determination of water soluble extractive:

5 gram of air dried drug. coarsely powdered *Rasa Chenduram* was macerated with 100ml of distilled water in a closed flask for twenty-four hours, shaking frequently. The solution was filtered and 25 ml of filtered was evaporated in a tarred flat bottom shallow dish, further dried at 1000c and weighted. The percentage of water soluble extractive was calculated with reference to the air dried drugs.

### 2.6.7 Determination of alcohol soluble extractive:

2.5gram of air dried drugs coarsely powdered *Rasa Chenduram* was macerated with 50ml. alcohol in closed flask for 24 hours. With frequent shaking.it was filtered rapidly taking precaution against loss of alcohol .10ml of filtrate was the evaporated in a tarred flat bottom shallow dish, dried at 1000 c and weighed. The percentage of alcohol soluble extractive was calculated with reference to air dried drug.

### 2.6.8 Details regarding the FT-IR analysis.

FT-IR spectra were recorded at Tamilnadu test house Vanakaram, Chennai, India. The Perkin Elmer Spectrum One Fourier Transform Infrared (FTIR) Spectrometer was used to derive the FT IR Spectra of *Rasa chenduram* in Potassium Bromide (KBr) matrix with scan rate of 5 scan per minute at the resolution 4cm<sup>-1</sup> in the wave number region 450-4000cm<sup>-1</sup>.

## 3. Results and discussion:

**Table 1. Physical characterization of *Rasa Chenduram***

S.no	Parameter	Result
1	Colour	Reddish-brown in colour
2	State of the drug	Powder
3	Consistency	Fine powder
4	Solubility	Sparingly soluble in water, DMSO Well soluble in acid(HCL and H <sub>2</sub> SO <sub>4</sub> )

5	Sense of touch	Fine
6	Sense of taste	Tasteless
7	Sense of smell	No significant smell is observed

**Table 2. Result of Physico chemical parameters**

Sl.no	Parameters	Result
1	Specific gravity	0.967
2	Ph	4.76%
3	Particle size	Completely passes through sieve No.120
4	Total Ash	1.8%
5	Acid insoluble ash	Less than 1%
6	Water Soluble Ash	Less than 1%
7	Water Soluble Extractive	2.3%
8	Alcohol soluble Extractive	1.91%
9	Loss on drying at 105 degree Celsius	52.02%

### 3.1 Discussion on physic-chemical analysis

#### a) Solubility:

Solubility is one of the important parameters to attain the desired concentration of drug in systemic circulation and the required pharmacological response. *RC* is soluble in the major solvent, sparingly soluble in some of the solvents there yet proves its efficiency of solubility in the stomach indirectly, increased in bio- availability.

#### b) pH (potential hydrogen)

*Rasa chenduram* shows acidic pH. This pH level plays a role in enzyme activity by maintaining the internal environment thus regulating the homeostasis. It is also an important factor in drug absorption.

#### c) Specific gravity

The trial drug *RC* shows specific gravity which is lesser than water. It shows a nature of absorption.

#### d) Loss on drying

The low moisture content of *RC* indicates that it has a long shelf life. Moisture content increased can adversely affect the active ingredient. But, *RC* moisture doesn't damage it. So the low moisture content of *RC* offers maximum microbial stability.

#### e) Ash Values Total Ash value

The low total Ash value of *RC* indicates the richness of organic substances like Sodium, Chloride, Calcium, and Potassium. These organic compounds are responsible for the mineral supplements and therapeutic effect of *RC*.

#### f) Acid insoluble ash

Lowering the acid insoluble ash value better will be the drug quality. This drug possesses a low value (Less than 1%) of acid-insoluble ash indicating that the preparation did not contain any sand, dust or stones.

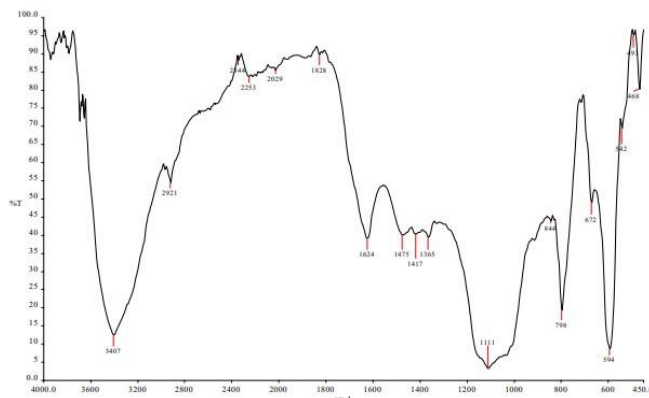
#### g) Water soluble ash

Decreased water-soluble ash value (Less than 1%) indicates easy facilitation of diffusion and osmosis mechanisms. This nature might be helpful for the better absorption

### 3.1 Discussion in FT-IR

FT-IR (Fourier Transform Infra-Red spectroscopy) FTIR instrumental analysis was done. The test drug was identified to have 12 peaks. They were the functional groups present in the trial drug *Rasa chenduram*. The Table 3 and Figure show the presence of Alcohol, Alkanes, Alkene, Phenols, Alkyl halide, aromatics, carboxyl, and Nitrile groups which represent the peak value. The FTIR analysis of *RC* shows the spectrum that appears which denotes the molecular absorption and transmission. It forms the molecular fingerprint of the sample. It is the functional group and determines the number of compounds present in the sample. These functional groups may be responsible for the therapeutic effect of the drug<sup>[5]</sup>

FT-IR (Fourier Transform Infra-Red)



**Table 3. Result of FT-IR**

Characteristic IR Absorption Frequencies of Organic Functional Groups			
Functional Group	Type of Vibration	Characteristic Absorptions (cm-1)	Intensity
Alcohol			
O-H	(stretch, H-bonded)	3407	<b>strong, broad</b>
Alkane			
-C-H	Bending	1365	<b>Variable</b>
Alkene			
=C-H	Bending	798	<b>Medium</b>
=C-H	Bending	844	<b>Strong</b>
C=C	Stretch	1624	<b>Variable</b>
Alkyl Halide			
C-Br	Stretch	594	<b>Strong</b>
C-Br	Stretch	542	<b>Strong</b>
Aromatic			
C-H	Stretch	1417	<b>medium-weak, multiple bands</b>
C-H	Stretch	1475	<b>medium-weak, multiple bands</b>
Carboxyl			
C=O	Stretch	1828	<b>Strong</b>
Nitrile			
CN	Stretch	2253	<b>Medium</b>
Carboxyl			
OH-C-O	Stretch	2344	<b>strong, two bands</b>
Phenols			
	O-H stretch	3644	<b>strong</b>

The sophisticated instrumental analysis like FTIR shows the presence of functional groups through their stretch and bends which are responsible for its functional activity. scrutinizing all the above studies, it is concluded that the Siddha herbal formulation *Rasa Chenduram* was subjected to many studies to validate its efficacy and safety through proper standardization procedure and the results revealed its potency and efficacy. The wavenumber from 1500 cm-1 to 400 cm-1 provides particulars about the molecular fingerprint.



- The above result showed the presence of functional groups like primary and secondary alcohol, phenols, alkanes, and alkyl halides in “*Rasa Chenduram*”.
- They may be responsible for the presence of anticancer action of *RC* in cancer.

#### **Phenols :**

- The effect of phenols is currently of great awareness due to their anti-oxidative and possible anti-carcinogenic activities.
- Phenols and flavonoids possess diverse biological activities, for example, antiulcer, antioxidant, cytotoxic and antitumor, anti-inflammatory, antispasmodic and antidepressant activities.<sup>[6]</sup>

#### **Alcohol:**

- OH group of *CMC* has a higher potential for inhibitory activity against microorganisms.<sup>[7]</sup>

#### **Alkyl halide:**

- High proportion of low molecular weight alkyl halides may be weakly carcinogenic and provide evidence supporting an electrophilic hypothesis of carcinogenesis<sup>[8]</sup>.

#### **Alkanes :**

Alkane derivative possesses anti-cancer activity. Through the observed frequencies, because of this reason though Siddha literature states that this drug is indicated for diseases such as Cancer, Leprosy, Vatha, Syphilis etc<sup>[9]</sup>. So, the author hopes that this study could help future studies regarding “*Rasa Chenduram*”

#### **Carboxylic acid**

Carboxylic acid Benzene-poly-carboxylic Acid Complex (BP-CI) is a novel anticancer complex against human cancer cells. Docosahexaenoic acid (DHA) is an omega-3 fatty acid. Its structure is a carboxylic acid (-oic acid) with a 22- carbon chain (docosa-is Greek for 22) and six (hexa-) cis double bounds.. In mice, DHA was found to reduce growth of human colon carcinoma cells The cytotoxic effect of DHA was caused by decrease in cell growth regulator<sup>[10]</sup>

#### 4. Conclusion

The purification process takes many steps to attain a non-toxic form. In this study, the higher-order Siddha formulation *Rasa chenduram* undergoes for purification process to attain the GMP. The above report could be used as a finger print for future references in standardization of “*Rasa Chenduram*” evidenced based data provide valuable information to the researchers working on this formulation in near future.

#### 5. Reference:

1. Khan TA, Gohel AK, Mallya R. Standardisation and HPTLC method development of marketed Ayurvedic formulation-balarishta. *Int J Pharm Pharm Sci* 2016;8:201-9
2. Abinaya r. karolindaisy rani .r scientific validation of anti-oral cancer, anti-tumour and anti-microbial activities of siddha metallomineral formulation “ kaalamega narayana chendhooram ” in in-vitro studies. *WJPR* volume 8, issue 9, 789-811.
3. Dr. Thiagarajan, Siddha Material Medica (mineral and animal kingdom), First edition, 2008, Translation and publication wing, Dept of Indian Medicine and Homoeopathy.pg.no244,305, 325.
4. WHO GUIDELINES
5. FTIR Spectroscopy available at:[https://www.lpdlabsservices.co.uk/analytical\\_techniques/chemical\\_anaysis\\_ftir.php](https://www.lpdlabsservices.co.uk/analytical_techniques/chemical_anaysis_ftir.php)
6. Malinee Pongsavee. Effect of borax on immune cell proliferation and sister chromatid exchange in human chromosomes. US National library of medicine.
7. Jay A. Young. Potassium Nitrate. *The journal of chemical foundation*. DOI: 10.1021/ed082p1305; September 1, 2005; *J. Chem. Educ.*, 2005, 82 (9), p 1305. Available at: <https://cdn-pubs.acs.org/doi/10.1021/ed082p1305>.
8. F. S. Williamson Basic Copper Sulphate, *The journal of physical chemistry*. ACS Publications; DOI:10.1021/j150233a005; January 1922. Pg.789-797, Available at <http://pubs.acs.org/doi/abs/10.1021/j150233a005>
9. Malinee Pongsavee. Effect of borax on immune cell proliferation and sister chromatid exchange in human chromosomes. US National library of medicine,
10. Jay A. Young. Sodium Chloride. *The journal of chemical foundation*. ACS Publications; DOI: 10.1021/ed084p1758; November 1, *Educ.*, 2007, 84 (11), p 1758 2007; *J. Chem.*