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Research Article

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Physicochemical analysis of Eraippu Noi Chooranam, A Siddha polyherbal formulation

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Keywords

Poly Herbal formulation, High Performance Thin Layer Chromatography, Physico chemical Analysis, Phytochemistry, Siddha.

Abstract

Background: Standardization of Siddda drugs is the need of the hour. Eraippu Noi Chooranam is a modified Siddha Poly Herbal formulation indicated for respiratory diseases in the text Siddha research pharmacopoeia. **Aim and Objective**: To do physicochemical analysis for the drug Eraippu Noi Chooranam. **Materials and Methods:** The drug is prepared as per the method mentioned in the classic siddha literature. The drug is subjected to physicochemical analysis such as total ash, loss on drying, total sugar, reducing sugar, fat content, microbial load, heavy metal analysis , Thin Layer Chromatography and High Performance Thin Layer Chromatography as per the Pharmacopeial laboratory standards of Indian medicine. **Results and Conclusion**: The drug is free of microbial contamination , Aflatoxins and Pesticide Residues. The heavy metals such as arsenic, mercury, cadmiumand lead are not detected.

1. Introduction

The Siddha system of medicine is predominantly practiced in South India. While accepting its benefits global community demands evidence based scientific explanation to understand the concept of Siddha system of medicine and demands quality matching International standards to reassure the efficacy of Siddha medicine. *Eraippu Noi Chooranam* is a classic Siddha drug chosen from the text *Siddha Research Pharmacopoeia*. It is indicated for asthma, chronic bronchitis and flatulence[1]. The use of scientific tools are essential to validate the traditional claim. Though Siddha drugs are considered to be safe and effective, it is the utmost duty of the physicians to standardize the formulation before trying out in humans. The drug is a poly herbal drug and all the ingredients included are very effective in curing kapha diseases. The main aim of this study is to evaluate the physico chemical characters of the drug *Eraippu Noi Chooranam* .

2. Aim and objectives

The aim of this study is to do phsico chemical analysis , HPTLC finger printing for the drug *Eraippu Noi Chooranam*

3. Materials and Methods

3.1Collection and Identification of plant materials

The herbal ingredients were authenticated by the Assistant Professor of Medicinal botany , National Institute of Siddha, Chennai. The raw drugs were purified as per the methods mentioned in the literature.

3.2. Preparation of the drug *Eraippu Noi Chooranam* [2]:

Ingredients:

Kuppaimeni leaves choornam (*Acalypha indica*)	- 224 gms
Chiru Cherupadai leaves choornam (*Mollugo lotoides*)	- 224 gms
Potrilai kaiyan leaves choornam (*Eclipta prostrata*)	- 224 gms
Vembu leaves choornam (*Azadiracta indica*)	- 224 gms
Milagu fried chooranam (*Piper nigrum*)	- 112 gms
Arisi thippili chooranam (*Piper longum*)	- 112 gms
Amukkara chooranam (*Withania somnifera*)	- 112 gms
Kadukkai thol chooranam (*Terminalia chebula*)	- 112 gms
Cane sugar powder	-392 gms

3.3. Purification of raw drugs[3,4]:

The raw drugs are purified as per the methods mentioned in the Siddha literatures .

3.4. Analytical specifications of semisolid drugs [5] :

1..Description : Particle size 2. Loss on drying at 1050 C, 3. Total – ash, 4. Acid – insoluble ash, 5. pH, 6. Total solid,7. Reducing sugar,9. Total sugar, 10. Identifications : TLC/HPTLC, 11. Test for heavy metals: Lead, Cadmium,Mercury, Arsenic, 12. Microbial contamination: Total bacterial count,Total fungal count, 13. Test for specific Pathogen: E. coli, Salmonella spp., S.aureus, Pseudomonas aeruginosa,14. Pesticide residue: Organochlorine pesticides, Organophosphorus pesticides, 15 Test for Aflatoxins (B1,B2,G1,G2).

3.5 HPTLC Analysis of aqueous extract of ENC

Materials and methods

Test solution preparation

The ENC sample of 1g was weighed in an electronic balance and dissolved in 10ml of Aqueous solvent and centrifuged at 3000rpm for 5min. respectively. These solutions were used as test solution for HPTLC analysis.

Sample application

10µl of test solutions and 5µl of standard solution were loaded as 6mm band length in the 10 x 10 Silica gel 60F_{254}TLC plate using Hamilton syringe and CAMAG LINOMAT 5 instrument.

Spot development

The samples loaded plate was kept in TLC twin trough developing chamber (after saturated with Solvent vapor) with respective mobile phase (standards) and the plate was developed in the respective mobile phase up to 80mm.

Photo-documentation

The developed plate was dried by hot air to evaporate solvents from the plate. The plate was Photodocumented the images at UV 254nm.

3.6. Scanning

The plate was fixed in scanner stage (CAMAG TLC SCANNER 3) and scanning was done at UV 254nm. The Peak table, Peak display and Peak densitogram were noted. The software used was winCATS 1.3.4 version.

3.7. Physicochemical analysis[5]:

The sample is tested for the following parameters as per the guidelines followed by WHO. Loss on drying, Total ash, Acid insoluble ash, Water soluble extractive, Alcohol soluble extractive, Particle size, Microbial load, Aflatoxins and Heavy metals.

4. Results and Discussion

4.1.Organoleptic characters:

Colour- brown.
Odour–Typical
Taste : sweet Consistency- powder

4.2. Physico-Chemical Parameters:

The results of the physicochemical parameters are given in Table 1.

Table 1. physicochemical parameters

Sl. No	Parameters	Result of Eraippu Noi Chooranam
1	Loss on drying at 105 degrees	3.77 %
2	Total- Ash	4.07 %
3	Acid- Insoluble	0.72%
4	Water- soluble extractive	68.03%
5	Alcohol- soluble extractive	24.97%
6	Particle size	
6.1	Material passing through ASTM 80(180 micron)	83.17%
6.2	Material passing through ASTM 80(180 micron)	79.33%

Loss on drying indicates the moisture content . The total ash content is the measure of inorganic constituents present in the drug. High ash content explains its unsuitable nature to be used as drug.

4.3. Heavy metal analysis using ICPOES:

The observed results of heavy metal analysis is tabulated below in table 2.

Table 2: Results of heavy metal analysis:

Heavy Metal	Specification as per AYUSH/WHO/FDA(26….)	Observed Result
Lead	10ppm	ND
Arsenic	3.0ppm	ND
Cadmium	0.3ppm	ND
Mercury	1ppm	ND

The heavy metals such as lead cadmium, arsenic and mercury are not detected

4.4. Test for Aflatoxins and Pesticide Residues:

Table 3: Results of test for Aflatoxins and Pesticide Residues**:**

Test	Observed Result
Aflatoxin B1	4.06µg/kg
Aflatoxin B2	ND
Aflatoxin G1	1.34µg/kg
Aflatoxin G2	ND
Organophosphorus	ND
Organochloride	ND
Synthetic pyrethroids	ND

4.5. Test for Bacterial and Fungal count[6]:

Table 4: Results of test for bacterial and fungal count:

Test	Specification as per	Observed Result
	AYUSH/WHO/FDA	
Total bacterial count	NMT 105 CFU/g	18,560CFU/g
Total fungal count	NMT 103 CFU/g	< 10 CFU/g
E.coli	Absent/g	Absent/g
Salmonella	Absent/g	Absent/g
Pseudomonas aeruginosa	Absent/g	Absent/g
Staphyloccus aureus	Absent/g	Absent/g

The bacterial and fungal load are within the prescribed limits. The above results suggests that the prepared drug Eraippu Noi Chooranam is of standard quality.

A HPTLC profile is done for the drug Eraippu Noi Chooranam. The HPTLC image is shown in Figure1.

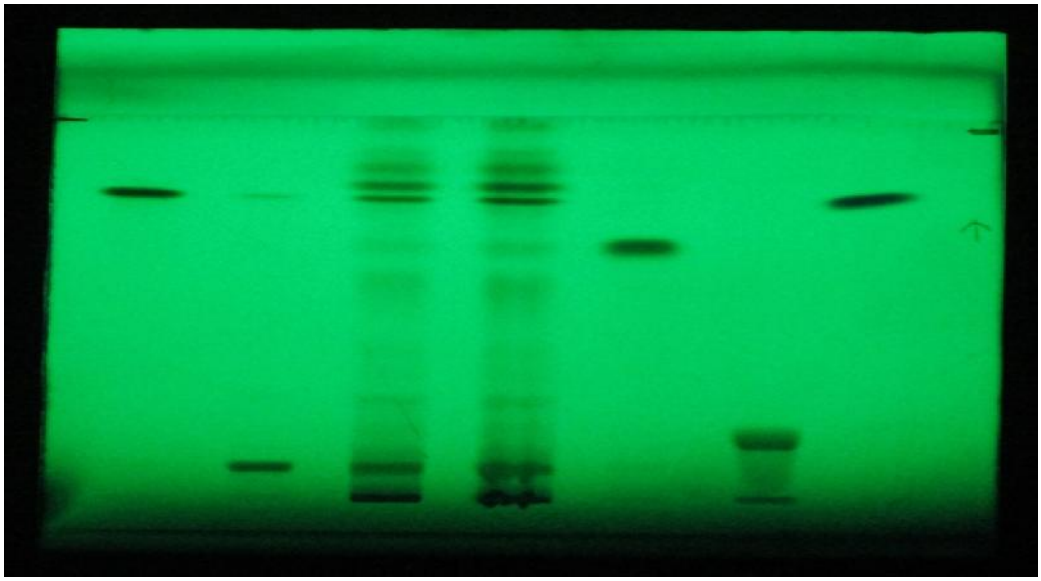
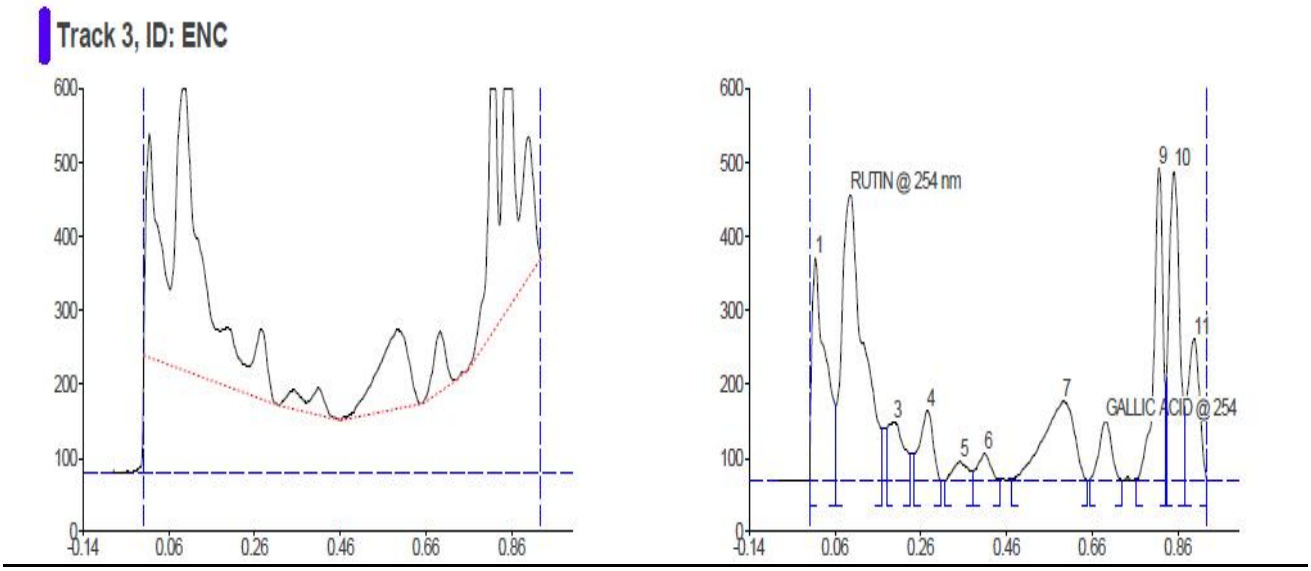


Fig 1 HPTLC chromatogram of standards and SRB

Track 1 Quercetin - standard as reference marker
Track 2 Rutin standard as reference marker
Track 3 Aqueous extract of ENC
Track 4 Aqueous extract of ENC
Track 5 Gallic acid - standard as reference marker
Track 6 Mangiferin- standard as reference marker
Track 7 Cafffeic acid-standard as reference marker



Peak table

Track No	Amount of Sample	Peak	Rf	Name of std
L	5µl	**1**	**0.87**	**Quercetin**
2	5 µl	**1**	0.09	**Rutin**
3	10 µl	**1**	0.01,**0.09**, 0.2,0.27,0.35,0.41,0.59,**0.69,**0.82**,0.85,0.90**	**Rutin and gallic Present**
4	10 µl	**1**	0.01,0.10,0.20,0.27,0.35,0.41,0.58,0.69,**0.81**,0.85,0.90	**Quercetin**
5	5 µl	**1**	**0.69**	**Gallic acid**
6	5µl	**8**	0.16	**mangiferin**
7	**5**µl	**8**	0.83	**Caffeic acid**

Report

By comparing with flavonoid standards Gallic acid and RUTIN , the ENC sample exactly match with Rf value and it was confirmed that it contains rutin and gallic acid
Also considered to be may be presence of caffeic acid.

Conclusion

Based on the above results , it can be assumed that the drug Eraippu Noi Chooranam has validated the traditional claim

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