International Journal of Current Advanced Research
ISSN: O: 2319-6475, ISSN: P: 2319-6505, Impact Factor: 6.614
Available Online at www.journalijcar.org
Volume 10; Issue 06 (D); June 2021; Page No.24682-24686
DOI: http://dx.doi.org/10.24327/ijcar.2021.4918.24686

Research Article

STANDARDIZATION OF MĀTUḶAI NEY: AN HERBAL GHEE BASED SIDDHA MEDICINAL PREPARATION

Prabhavathi N., Thiruveni K and Shyamala K

Department of KuzhanthaiMarunthuvam, Govt. Siddha Medical and Hospital, Palayamkottai

A R T I C L E I N F O

Article History:

Received 13th March, 2021
Received in revised form 11th April, 2021
Accepted 8th May, 2021
Published online 28th June, 2021

Key Words:

Siddha medicine, standardization, PLIM, organoleptic characters, physico-chemical characters, phytochemicals

A B S T R A C T

Standardization of herbal and herbo-mineral drugs refers to confirmation of identify and determination of its quality, purity and detection of adulteration by various parameters like morphological, microscopical, physical, chemical and biological observations. As per Pharmacopeial Laboratory of Indian Medicine (PLIM) guidelines there are some parameters to follow the standardization techniques to improve the efficacy of Siddha drugs. As a consequence of focusing upon the current need of drug standardization the present work was undertaken to standardize the traditional Siddha formulation *mātuḷainey* as per PLIM guidelines given for*ney*.

INTRODUCTION

Standardization of herbal and herbo-mineral drugs refers to confirmation of identify and determination of its quality, purity and detection of adulteration by various parameters like morphological, microscopical, physical, chemical and biological observations. Quality assurance is a system for performance, service of the quality of a product against system, standard in specified requirement for the people. As Siddha system perpetuating for centuries so many traditional Siddha formulations are still not yet standardized. Only few standard tests are given for some preparations of herbal and herbo mineral drugs in Siddha literatures (ex. A well finished parpam when pour in water it won't soak and float in its surface). Preparation method of siddha drugs varies from person to person. So, yet there is no standard method was fixed. Hence, considering the global need standardization of Siddha drugs becomes highly potential in order to explore its potency and efficacy in global market. As per Pharmacopeial Laboratory of Indian Medicine (PLIM) guidelinesthere are some parameters to follow the standardization techniques to improve the efficacy of Siddha drugs. As a consequence of focusing upon the current need of drug standardization the present work was undertaken to standardize the traditional Siddha formulation *mātuḷainey* as per PLIM guidelines given for *ney*.

Corresponding author:* **Prabhavathi N
Department of KuzhanthaiMarunthuvam, Govt. Siddha Medical and Hospital, Palayamkottai

MATERIALS AND METHODS

Drug selection
The trial drug *mātuḷainey is* selected from Piḷḷaippiṇimaruttuvam part-II–Department of Indian medicine and homeopathy, Chennai-106, 2006. for the management of *CūliKaṇam*

Authentication of raw material

This will be performed by geologists in case of Metal/ Mineral ingredients and by a taxonomist in case of herbal ingredients.

Purification of raw material

Purification process will be performed as per theclassical siddha texts.
Preparation
mātuḷaicamūlaccāṟu - 325ml
cīntilcarkkarai - 90 gram

These 2 drugs are taken in an earthen pot & kept under the flame & heated till it attains the *kuḻampu* consistency. To this 325ml of cow's butter is added.
cātikkāy - 18 gm
cātipattiri - 18 gm
ēlam - 18 gm

After this the powdered form of above three is added and subjected to heat and kept in flame till it attains the *ney* consistency.

Administration of the drug

Dosage: *5*-10 ml (twice a day)

Duration: 41days.

Authentication of final drug

The final product was authenticated by the trained experts from Gunapadam department of Govt. Siddha medical college and hospital, Palayamkottai, Tirunelveli- 627002 for its completion.

Quality assurance of the prepared drug
Quality assurance was performed as per the PLIM guidelines and the analytical parameters done as follows.
Physico- chemical evaluation

Determination of Iodine value

About 20 gm of test sample was transferred into Iodine flask. To which 10 ml of chloroform was added and warmed slightly and cooled for 10 minutes.

Followed by this about 25 ml of Wiji's solution was added in the same flask and shaken well. The flask was allowed to stand for 30 mins and refrigerated for an hour. T About 10 ml of KI solution was added to this and titrated against 0.1N Sodium thiosulphate solutions until the appearance of yellow colour. 1 ml of starch indicator was added and again titrated against the sodium thiosulphate solution from the burette. Disappearance of blue colour indicates end point. Repeat the above procedure without taking sample and note the corresponding reading for blank titration.

Determination of saponification value

About 2 gm of test sample was transferred into the round bottomed flask. To this about 20 ml of 0.5 N alcoholic KOH solutions was added to the round bottomed flask. Repeat the same procedure without taking the sample for blank titration. Reflux both sample and blank round bottomed flasks for 1 hour. After reflux, allow both the round bottomed flasks to cool. Titrate the samples using 0.5 N HCl with phenolphthalein indicator. The disappearance of pink indicates the end point.

Determination of Viscosity value

Viscosity determination were been carried out using Ostwald viscometers. Measurement of viscosity involves the determination of the time required for a given volume of liquid to flow through a capillary. The liquid is added to the viscometer, pulled into the upper reservoir by suction, and then allowed to drain by gravity back into the lower reservoir. The time that it takes for the liquid to pass between two etched marks, one above and one bellow the upper reservoir, is measured.

Determination of Refractive Index

Determination of RL was carried out using Refract meter.

Determination of Weight per ml

Weight per ml was determined using the comparative weight calibration method, in which the weight of 1ml of the base of the formulation was calculated and then weight of 1 ml of finished formulation were been calculated. The difference between weight variations of the base with respect to finished formulation calculated as an index of weight per ml.

Acid Value

Accurately 5 g of test sample was weighed and transferred into a 250 mL conical flask. To this, a 50 mL of neutralized alcohol solution was added. This mixture was heated for 10 min by heating mantle. Afterwards, the solution was taken out after 10 min and 1 or 2 drops of phenolphthalein indicator was added. This solution was titrated against KOH solution from the burette. The appearance of pink colour indicated the end point. The volume of consumed KOH solution was determined and the titration of test sample was carried out in triplicate and the mean of the successive readings was used to calculate the acid-value of the respective sample by following expression.

Acid value = Titter Value X 0.00561X 1000 / Wt. of test sample (g) Peroxide value

5 g of the substance being examined, accurately weighed, into a 250-ml glass-stoppered conical flask, add 30 ml of a mixture of 3 volumes of glacial acetic acid and 2 volumes of chloroform, swirl until dissolved and add 0.5ml volumes of saturated potassium iodide solution. Allow to stand for exactly 1 minute, with occasional shaking, add 30 ml of water and titrate gradually, with continuous and vigorous shaking, with0.01M sodium thiosulphate until the yellow colour almost disappears. Add 0.5 ml of starch solution and continue the titration, shaking vigorously until the blue colour just disappears (aml). Repeat the operation omitting the substance being examined (b ml). The volume of 0.01Msodium thiosulphate in the blank determination must not exceed 0.1 ml.

Peroxide value = 10 (a-b)/w

Sterility test by pour plate method

Objective

The pour plate techniques were adopted to determine the sterility of the product.

Contaminated / un sterile sample (formulation) when come in contact with the nutrition rich medium it promotes the growth of the organism and after stipulated period of incubation the growth of the organism was identified by characteristic pattern of colonies. The colonies are referred to as Colony Forming Units (CFUs).

METHODOLOGY

Test sample was inoculated in sterile petri dish to which about 15 mL of molten agar 45oC were added. Agar and sample were mixed thoroughly by tilting and swirling the dish. Agar was allowed to completely gel without disturbing it. (about 10 minutes). Plates were then inverted and incubated at 37o C for 24-48 hours and further extended for 72 hrs for fungal growth observation. Grown colonies of organism was then counted and calculated for CFU.

TLC Analysis

Test sample was subjected to thin layer chromatography (TLC) as per conventional one-dimensional ascending method using silica gel 60F254, 7X6 cm (Merck) were cut with ordinary household scissors. Plate markings were made with soft pencil. Micro pipette was used to spot the sample for TLC applied sample volume 10- micro litre by using pipette at distance of 1 cm at 5 tracks. In the twin trough chamber with the specified solvent system After the run plates are dried and was observed

using visible light Shortwave UV light 254nm and light long-wave UV light 365 nm

High Performance Thin Layer Chromatography Analysis

HPTLC method is a modern sophisticated and automated separation technique derived from TLC. Pre-coated HPTLC graded plates and auto sampler was used to achieve precision, sensitive, significant separation both qualitatively and quantitatively. High performance thin layer chromatography (HPTLC) is a valuable quality assessment tool for the evaluation of botanical materials efficiently and cost effectively. HPTLC method offers high degree of selectivity, sensitivity and rapidity combined with single-step sample preparation. Thus, this method can be conveniently adopted for routine quality control analysis. It provides chromatographic fingerprint of phytochemicals which is suitable for confirming the identity and purity of Phyto therapeutics.

Chromatogram Development

It was carried out in CAMAG Twin Trough chambers. Sample elution was carried out according to the adsorption capability of the component to be analysed. After elution, plates were taken out of the chamber and dried.

Pesticides residues test

Extraction

Test sample were extracted with 100 ml of acetone and followed by homogenization for brief period. Further filtration was allowed and subsequent addition of acetone to the test mixture. Heating of test sample was performed using a rotary evaporator at a temperature not exceeding 40ºC until the solvent has almost completely evaporated. To the residue add a few millilitres of toluene and heat again until the acetone is completely removed. Resultant residue will be dissolved using toluene and filtered through membrane filter.
Heavy metal analysisby AAS
Standard: Hg, As, Pb and Cd – Sigma

METHODOLOGY

Atomic Absorption Spectrometry (AAS) is a very common and reliable technique for detecting metals and metalloids in environmental samples. The total heavy metal content of the sample was performed by Atomic Absorption Spectrometry (AAS) Model AA 240 Series. In order to determination the heavy metals such as mercury, arsenic, lead and cadmium concentrations in the test item.

Sample Digestion

Test sample was digested with 1mol/L HCl for determination of arsenic and mercury. Similarly, for the determination of lead and cadmium the sample were digested with 1mol/L of HNO3.

Standard reparation

As & Hg- 100 ppm sample in 1mol/L HCl
Cd & Pb- 100 ppm sample in 1mol/L HNO3
Aflatoxin Assay by TLC (B1,B2,G1,G2)

Solvent

Standard samples were dissolved in a mixture of chloroform and acetonitrile (9.8: 0.2) to obtain a solution having concentrations of 0.5 µg per ml each of aflatoxin B1 and aflatoxin G1 and 0.1 µg per ml each of aflatoxin B2 and aflatoxin G2.

Procedure

Standard aflatoxin was applied on to the surface to pre coated TLC plate in the volume of 2.5 µL, 5 µL, 7.5 µL and 10 µL. Similarly, the test sample was placed and Allow the spots to dry and develop the chromatogram in an unsaturated chamber containing a solvent system consisting of a mixture of chloroform, acetone and isopropyl alcohol (85: 10: 5) until the solvent front has moved not less than 15 cm from the origin. Remove the plate from the developing chamber, mark the solvent from and allow the plate to air-dry. Locate the spots on the plate by examination under UV light at 365 nm.

RESULTS AND DISCUSSION

Physico -chemical evaluation

Table 0 1 Physico -chemical evaluation

State	Semisolid
Nature	Dense viscous
Odor	Characteristic
Touch / Consistency	Greasy
Flow Property	Free flowing
Appearance	Intense yellow

Solubility Profile

Table 0 2 Solubility Profile

S. No	Solvent Used	Solubility / Dispersibility
1	Chloroform	Soluble
2	Ethanol	Insoluble
3	Water	Insoluble
4	Ethyl acetate	Soluble
5	DMSO	Insoluble

Analytical Report

Table 0 3 Analytical Report

S. No	Parameter	MN
1	Viscosity at 50°C (Pa s)	106.567
2	Refractive index	1.34
3	Weight per ml (gm/ml)	0.5315
4	Iodoine value (mg I2/g)	85.72
5	Saponification Value(mg of KOH to saponify 1gm of fat)	222.78
6	Acid Value mg KOH/g	0.7293
7	Peroxidase Value mEq/kg	2.42

Sterility test by pour plate method

Observation

No growth was observed after incubation period. Reveals the absence of specific pathogen

RESULT

No growth / colonies were observed in any of the plates inoculates with the test sample.

Table 0 4 Sterility test by pour plate method

Test	Result	Specification	As per AYUSH/WHO
Total Bacterial Count	Absent	NMT 10^5CFU/g	Asper AYUSH specification
Total Fungal Count	Absent	NMT 10^3CFU/g	

TLC Analysis

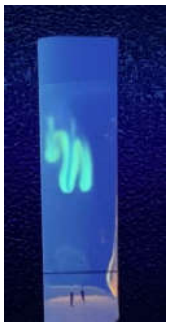

Figure 0 1 TLC Visualization of MN

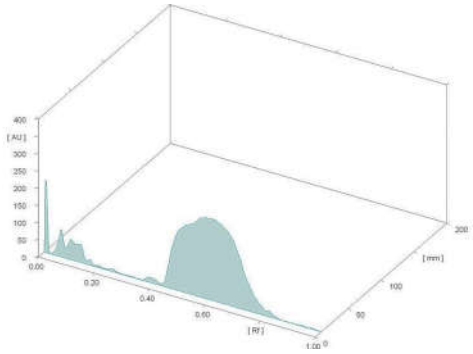

Figure 0 2 TLC plate visualization at 366 nm

High Performance Thin Layer Chromatography Analysis
HPTLC finger printing of Sample MN

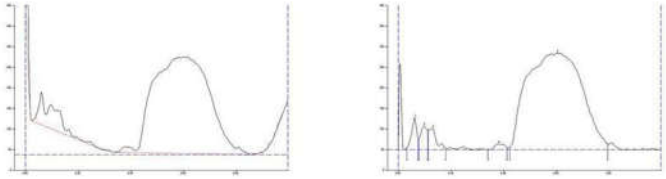

Figure 0 3 HPTLC finger printing of Sample MN

Peak Table

Table 0 5 HPTLC finger printing analysis

Peak	Start Rf	Start Height	Max Rf	Max Height	Max %	End Rf	End Height	Area	Area %
1	0.03	5.5	0.06	78.6	17.95	0.08	29.9	693.8	2.84
2	0.08	30.0	0.10	58.1	13.25	0.11	48.1	660.4	2.70
3	0.11	48.6	0.13	52.6	12.00	0.18	3.7	691.2	2.83
4	0.34	1.1	0.38	14.3	3.25	0.41	4.9	243.1	0.99
5	0.42	5.9	0.60	234.6	53.54	0.80	15.3	22147.8	90.63

Report

HPTLC finger printing analysis of the sample reveals the presence of five prominent peaks corresponds to presence of five versatile phytocomponents present with in it. Rf value of the peaks ranges from 0.03 to 0.42.

Pesticides residues test

Test Result Analysis of the Sample MN

Table 0 6 Pesticides residues test

Pesticide Residue	Sample MN	AYUSH Limit (mg/kg)
I.Organo Chlorine Pesticides		
Alpha BHC	BQL	0.1mg/kg
Beta BHC	BQL	0.1mg/kg
Gamma BHC	BQL	0.1mg/kg
Delta BHC	BQL	0.1mg/kg
DDT	BQL	1mg/kg
Endosulphan	BQL	3mg/kg
II.Organo Phosphorus Pesticides		
Malathion	BQL	1mg/kg
Chlorpyriphos	BQL	0.2 mg/kg
Dichlorvos	BQL	1mg/kg
III. Organo carbamates		
Carbofuran	BQL	0.1mg/kg
III.Pyrethroid		
Cypermethrin	BQL	1mg/kg

BQL- Below Quantification Limit

Result: The results showed that there were no traces of pesticides residues such as Organo chlorine, Organo phosphorus, Organo carbamates and pyrethroids in the sample provided for analysis

Heavy metal analysis by ASS

Table 0 7 Heavy metal analysis by ASS

Name of the heavy metals	Absorption max A max	Result analysis
Lead	217.0nm	31.5
Arsenic	193.7nm	10.98
Cadmium	228.8nm	3.16
Mercury	253.7nm	BDL

BDL- Below Detection Limit

Aflatoxin Assay by TLC (B1,B2,G1,G2)

Table 0 8 Aflatoxin Assay by TLC (B1, B2, G1, G2)

Aflatoxin	Sample MN	AYUSH Specification limit
B1	Not detected- absent	0.5ppm
B2	Not detected- absent	0.1ppm
G1	Not detected- absent	0.5ppm
G2	Not detected- absent	0.1ppm

RESULT

The results shown that there were no spots were being identified in the test sample loaded on TLC plates when compare to the standard which indicates that the sample were free from Aflatoxin B1, Aflatoxin B2, Aflatoxin G1, Aflatoxin G2.

CONCLUSION

In this study, it has been concluded that the trial drug *mātuḷainey* has been standardized using diverse scientific quality parameters. The analysis of the *mātuḷainey* revealed that the drug was prepared in a satisfactory manner for the objectives to which is recommended.

References

Piḷḷaippiṇimaruttuvam part-II–Department of Indian medicine and homeopathy, Chennai-106, 2006

Dr. D. R. Lohar MSc., PhD, Protocol for testing of Ayurvedic, Siddha and Unani medicines, Govt. of India, Department of AYUSH, Ministry of health and family welfare, Pharmacopoeial laboratory for Indian medicines, Ghaziabad.

India Pharmacopeia I Volume I, Government of India, Ministry of Health and Family welfare, Indian Pharmacopeia commission, 2014.

Pharmacopoeial Laboratory for Indian Medicine (PLIM) Guideline for standardization and evaluation of indian medicine which include drugs of Ayurveda, Unani and Siddha systems. Department AYUSH Ministry of Health & Family Welfare, Govt. of India

Indian standard methods of sampling and test for oils and fats Indian standard institution New Delhi 47-50. 1964

Luciana de CASTRO. Determining Aflatoxins B1, B2, G1 and G2 in Maize Using Florisil Clean Up with Thin Layer Chromatography and Visual and Densitometric Quantification. Ciênc. Tecnol. Aliment. vol.21 no.1 Campinas. 2001.

WHO guideline for assessing the quality of herbal medicines with reference to contaminants and residues. WHO Geneva. 2007.

Lohar. D.R. Protocol for testing of ASU medicines. Pharmacopeial Laboratory for Indian Medicines. Ministry of AYUSH. 2007.

How to cite this article:

Prabhavathi N *et al* (2021) 'Standardization of Mātuḷai Ney: An Herbal Ghee Based Siddha Medicinal Preparation', *International Journal of Current Advanced Research,* 10(06), pp. 24682-24686.
DOI: http://dx.doi.org/10.24327/ijcar.2021.4918.24686

****