

Standardization and Physio-chemical analysis of Varma polyherbal formulation Ualignai Kiyazham

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Abstract

Introduction: Siddha system of medicine is a traditional system of medicine that originated over a millineum years ago in Tamil Nadu. The treatment modality involves healing the mind, body and soul rather than curing the illness. Varmam is the branch of siddha system of medicine. Varma maruthuvam is based on the energy flow that flows throughout the body. Most of the varma medicines are effective in curing the disease but they have not been standardized yet. Standardization of Siddha Varma Polyherbal formulation becomes essential to establish the monograph of the particular formulation along with this, it encompasses the Genuity, purity and safety of the preparations intended for usage in patients. “ULIGNAI KIYAZHAM (UK)”, a polyherbal formulation in varma medicine, is traditionally used to varma kaayam (pain), thodu varmangal, padu varmangal, thatu varmangal. **Aim and objective:** To standardize “ULIGNAI KIYAZHAM (UK)”, a siddha varma polyherbal formulation using physiochemical analysis and other analytical techniques such as HPTLC and TLC, Heavy metal analysis. **Materials and methods:** Varma polyherbal formulation “Ualignai kiyazham (UK)”, drug mentioned in the textbook of “Formulary of Varma medicine (Varma marunthu sei muraigal 5th edition)” which is used to treat varma kaayam (pain). A scientific approach was undertaken to standardize the varma polyherbal formulation UK by conducting a through physiochemical evaluation, HPTLC and TLC thereby ensuring the establishment of its quality and efficacy parameters. **Results:** The results obtained from physicochemical evaluation shows that the Total ash value of Ualignai kiyazham (UK) was “8.42%”, in which the acid insoluble ash was “1.71”, loss on drying at 105°C of the formulation UK was noted to be “12.01%” in which water soluble extract value and alcohol soluble extract value was “7.93%”, and “1.21%” respectively. The pH value of the drug 5.62 which is indicates it is acidic in nature. HPTLC finger printing analysis of the sample reveals the presence of nine prominent peaks corresponds to the presence of nine components present within it. Heavy metal analysis by AAS result shows no traces of heavy metals such as Mercury, Arsenic, Lead and Cadmium. Microscopic observation of the particle size analysis reveals that the average particle size of UK was found to be 112.9±32.5 µm. **Conclusion:** The preliminary physiochemical analysis of the siddha drug UK provides a foundational fingerprint for future clinical studies, thereby facilitating the establishment of its safety, efficacy and quality parameters.

Keywords: Ualignai kiyazham, UK, Varma polyherbal formulation, Physiochemical analysis, HPTLC.

Introduction

Varmam is the branch of siddha system of medicine. Varma maruthuvam is based on the energy flow that flows throughout the body. The foundation of life energy is called Vaasi or Pranana. The place where the pranana resides are called as Varmam points. The total number of Varmam points in human body are more than a thousand among which 108 are important points (Padu varmam 12, Thodu varmam 96). Many points are bilateral points lying in both the halves of the body, others which lie in the axis are unpaired points. Siddha Varmam is non - invasive and time saving. Varmam is the trademark of Siddha Medicine based on vital energy circulation in our body. Any disturbance in the flow of this energy leads to disease. Varmam Points are the places where this vital energy resides and activates both body and mind. By the proper stimulation of Varmam Points, restoration of normal function of body is possible. Varmam treatment includes two main elements varmam

physical manipulation and varmam medicines (Kiyazham, Ennai, Chooranam, Nei/Gritham.etc). Herbal drugs have been used widely because of its availability, cost effectiveness and safer than other drugs. Varma drugs requires standardization for global acceptance. Thus, the author preferred to choose Varma drug UK for standardization. It was aimed to validate the qualitative and quantitative screening for UK scientifically to prevent adulterations. Thus, Organoleptic properties, Physio-chemical parameters, HPTLC finger printing aspects according to PLIM guidelines, Heavy metal analysis by AAS were carried out in this drug.

Materials and Methods

Selection of the trial drug: In this research paper purified and prepared “UK” was taken as a trial drug for Lumbar spondylosis from the varma literature “Formulary of Varma medicine (Varma marunthu sei muraigal 5th edition pg. no-57)”

Table No1: Ingredients with Botanical name of ULIGNAI KIYAZHAM (UK)

S.no	Tamil name	Botanical name	Family	Parts used	Quantity
1	Ulinai	<i>Cardiospermum halicacabum</i>	Sapindaceae	Root	1 part
2	Thulasi	<i>Ocimum sanctum</i>	Lamiaceae	Root	1 part
3	Nannari	<i>Hemidesmus indicus</i>	Asclepiaceae	Root	1 part
4	Narchangan	<i>Clerodendrum inerme</i>	Verbenaceae	Root	1 part
5	Seendhil	<i>Tinospora cordifolia</i>	Menispermaceae	Root	1 part
6	Kurunthotti	<i>Sida cordifolia</i>	Malvaceae	Root	1 part
7	Thaneervittan	<i>Asparagus racemosus</i>	Liliaceae	Rhizome	1 part
8	Vilvam	<i>Aegle marmelos</i>	Rutaceae	Root	1 part
9	Kandankathiri	<i>Solanum xanthocarpum</i>	Solanaceae	Root	1 part
10	Adathodai	<i>Adathoda vasica</i>	Acanthaceae	Root	1 part
11	Muthakaasu	<i>Cyperus rotundus</i>	Cyperaceae	Root	1 part
12	Seeragam	<i>Cuminum cyminum</i>	Fabaceae	Seeds	½ part
13	Kothamalli	<i>Coriandrum sativum</i>	Apiaceae	Seeds	½ part

14	Maasikai	<i>Quercus infectoria</i>	Fabaceae	Fruit	½ part
15	Thippili	<i>Piper longum</i>	Piperaceae	Root	½ part
16	Chukku	<i>Zingiber officinale</i>	Zingiberaceae	Rhizome	½ part
17	Kostam	<i>Costus speciosus</i>	Costaceae	Rhizome	½ part
18	Elam	<i>Elettaria cardamomum</i>	Zingiberaceae	Seeds	½ part
19	Jathipathiri	<i>Myristica fragrans</i>	Myristicaceae	Aril(Seed coat)	½ part
20	Thalisapathiri	<i>Abies spectabilis</i>	Pinaceae	Leaves	½ part
21	Adhimathuram	<i>Glycyrriza glabra</i>	Fabaceae	Root	½ part
22	Thippili	<i>Piper longum</i>	Piperaceae	Root	½ part

Collection of the drug materials: The raw drugs was collected from MSS Aasaan and sons store Nagarcoil, Kanyakumari district, Tamilnadu.

Identification and authentication of the drugs: All the raw materials were identified and authenticated by the Botanical and Pharmacological experts at Government Siddha Medical College, Arumbakkam and Chennai.

Methods of purification: In UK all the drugs were dried and cleaned with white cloth.

Preparation of the drug: The Purified raw drugs viz *Cardiospermum halicacabum*, *Ocimum sanctum*, *Hemidesmus indicus*, *Clerodendrum inerme*, *Tinospora cordifolia*, *Sida cordifolia*, *Asparagus racemosus*, *Aegle marmelos*, *Solanum xanthocarpum*, *Justicia vasica*, *Cyperus rotundus*, *Cuminum cyminum* each 1 palam(35grams); *Coriandrum sativum*, *Quercus infectoria*, *Piper longum*, *Zingiber officinale*, *Costus speciosus*, *Elettaria cardamomum*, *Myristica fragrans*, *Abies spectabilis*, *Glycyrriza glabra*, *Piper longum* each half palam(17.5grams) are taken and ground into coarse powder using a stone mortar. To this 2 padi (2.6l) of water is added and mixed. This mixture is heated, until it gets reduced to 1/8 part to form Ulinai kiyazham. To this decoction, some amount of honey is added and taken.

Administration of the drug:

Form of the medicine: Kiyazham (decoction)
Route of administration: Oral (Internal)

Dose: 60ml twice a day depending on the severity
Shelf life: 3 hours.

Indication: *Ellaippu, koluthu, kuthulaichal, gunmam, soolai, vaatham, varmam theerum.*

Standardization of the drug

1. Organoleptic characters of UK:

State, nature, odour, touch, flow property, appearance of the drug was noted.

2. Physiochemical properties of UK:

A. Percentage Loss on Drying

Test drug was accurately weighed in evaporating dish. The sample was dried at 105°C for 5 hours and then weighed.

B. Determination of Total Ash

Test drug was accurately weighed in silica dish and incinerated at the furnace a temperature 400°C until it turns white in colour which indicates absence of carbon. Percentage of total ash will be calculated with reference to the weight of air-dried drug.

C. Determination of Acid Insoluble Ash

The ash obtained by total ash test will be boiled with 25 ml of dilute hydrochloric acid for 6mins. Then the insoluble matter is collected in crucible and will be washed with hot water and ignited to constant weight. Percentage of acid insoluble ash

will be calculated with reference to the weight of air-dried ash.

D. Determination of Alcohol Soluble Extractive

Test sample was macerated with 100 ml of Alcohol in a closed flask for twenty-four hours, shaking frequently during six hours and allowing it to stand for eighteen hours. Filter rapidly, taking precautions against loss of solvent, evaporate 25 ml of the filtrate to dryness in a tared flat bottomed shallow dish, and dry at 105°C, to constant weight and weigh. Calculate the percentage of alcohol-soluble extractive with reference to the air-dried drug.

E. Determination of Water Soluble Extractive

Test sample was macerated with 100 ml of chloroform water in a closed flask for twenty-four hours, shaking frequently during six hours and allowing it to stand and for eighteen hours.

Filter rapidly, taking precautions against loss of solvent, evaporate 25 ml of the filtrate to dryness in a tared flat bottomed shallow dish, and dry at 105°C, to constant weight and weigh. Calculate the percentage of water-soluble extractive with reference to the air-dried drug.

F. pH determination

Required quantity of test sample was admixed with distilled water and the subjected to screening using pH meter.

3. Heavy metal analysis by AAS

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

4. Particle size determined by Microscopic method

Particle size determination was carried out by the optical microscopic method. In which the sample was dissolved in sterile distilled water (app 1/100th dilution). Diluted samples were mounted on the slide and fixed with the stage of an appropriate location. Light microscopic images were drawn with a scale micrometer to arrive at the average particle size. Minimum of 30 observations were made to ascertain the mean average particle size of the sample.

5. Thin Layer Chromatography (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 were used to spot the sample for TLC applied sample volume 10-micro liter 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 Short-wave UV light 254nm and light long-wave UV light 365 nm

6. 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 phototherapeutics.

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.

Scanning: Plates were scanned under UV at 366nm. The data obtained from scanning were brought into integration through CAMAG software. Chromatographic finger print was developed for the detection of phytoconstituents present in each sample and their respective Rf values were tabulated.

Results

Physiochemical analysis

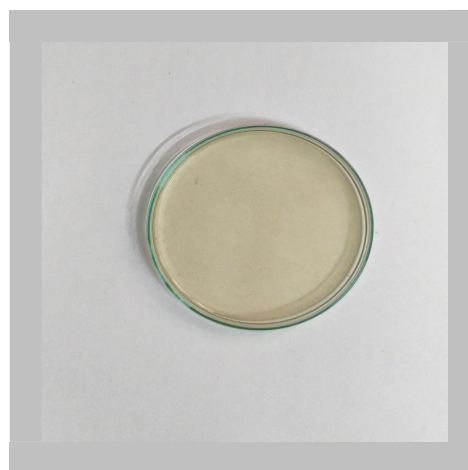


Table 2 Organoleptic character of UK

State	Solid	Liquid
Nature	Woody Fibrous	Non Viscous
Odour	Earthy	Mild odour
Touch	Hard	Non greasy
Flow Property	Non Free flowing	Free Flowing
Appearance	Light Brownish	Whitish

Table 3 Solubility Profile of UK

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

Table 4 Physiochemical evaluation of UK

S.No	Parameter	Mean (n=3) SD
1.	Loss on Drying at 105 °C (%)	5.62 %
2.	Total Ash (%)	8.42 %
3.	Acid insoluble Ash (%)	1.71 %
4.	Water soluble Extractive (%)	7.93 %
5.	Alcohol Soluble Extractive (%)	1.21 %
6.	pH	7.62

Heavy metal analysis by AAS**Table 5 Heavy metal analysis results of UK**

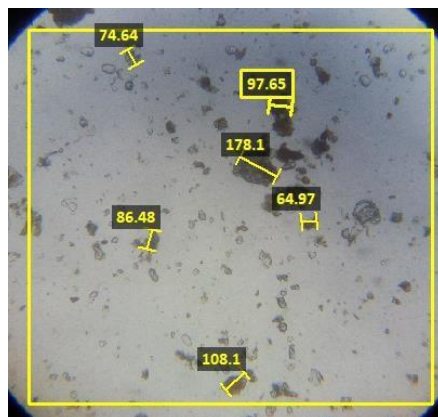
Name of the Heavy Metal	Absorption Max λ max	Result Analysis	Maximum Limit
Lead	217.0 nm	BDL	10 ppm
Arsenic	193.7 nm	BDL	3 ppm
Cadmium	228.8 nm	BDL	0.3 ppm
Mercury	253.7 nm	BDL	1 ppm

BDL- Below Detection Limit

Results of the present investigation have clearly shows that the sample has no traces of heavy metal such as Mercury, Arsenic, Lead and Cadmium.

Particle Size Determination by Microscopic Method

Microscopic Observation of Particle Size for the sample UK



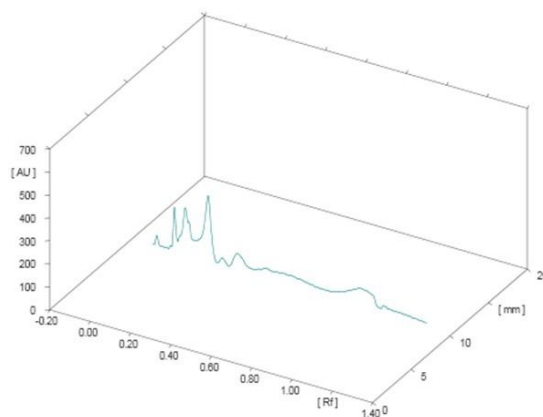
Report: Microscopic observation of the particle size analysis reveals that the average particle size of the sample was found to be $109.8 \pm 21.5\mu$.

HPTLC Report

TLC Visualization of UK at 366 nm



3D – Chromatogram



Discussion

The drug UK was coarsely powder and dark brownish colour. Fresh preparation of its extract shows non-Greasy, dark brownish with mild odour characteristic. Oral bio availability depends on several factors including aqueous solubility, drug permeability etc. The drug UK soluble in specific solvent like ethanol, water and DMSO nearby proves its efficiency of solubility increasing in bio available at in the stomach indirectly. The result derived from the physicochemical evaluation divulge that loss on drying at 105° C(%) value of UK was 5.62 which indicates the moderate moisture content could increase the stability and shelf life of the drug which is suitable for medicine preparation. Total Ash value (%) 8.42 which notes the presence of inorganic components. Water soluble extractive and alcohol soluble extractive value (%) was “7.93” and “1.21” respectively which indicates that the drug contains potential for better dissolution and absorption. The pH value of the drug was 7.6 which indicates that the drug is slightly alkaline and that the drug is well suited for most biological applications. Results of the heavy metal investigation have clearly shows that the sample has no traces of heavy metal such as Mercury, Arsenic, Lead and Cadmium. Microscopic observation of the particle size analysis reveals that the average particle size of UK was found to be $109.8 \pm 21.5 \mu\text{m}$. Rf value of the peaks ranges from 0.03 to 0.78. This study constitutes a preliminary exploration of UK physicochemical properties providing an essential foundation for future research which can build upon these insights to elucidate the complex characteristics of this varma polyherbal formulation. By expanding on this study discoveries, subsequent research can deeper into the therapeutic potential of UK, ultimately contributing to a more comprehensive understanding of its properties and applications in traditional medicines.

Conclusion

This study successfully standardized Ulinai kiyazham (UK) in accordance with PLIM guidelines and established protocols. The comprehensive standardization of this Siddha herbal formulation involved a range of parameters, including Organoleptic characters, Physicochemical parameters, TLC visualization at 366nm and HPTLC fingerprinting analysis. Results of the Heavy metal analysis have clearly shows that the sample has no traces of heavy metal such as Mercury, Arsenic, Lead and Cadmium. The results of this standardization effort will serve as a valuable tool for authenticating and evaluating the safety and quality of UK-I.

References

1. Dr. T. Kannan rajaram, Varma marunthu seimuraigal 5th edition pg.no- 57, 324
2. Dr. T. KannanRajaraam Varma Maruthuvam (Podhu).
3. Murugesu Mudhaliar, Gunapadam- Mooligai Vaguppu 9th edition, 2013
4. Xu Z. Particle and Size Distribution. Fundamentals of Air Cleaning Technology and Its Application in Cleanrooms. 2013;1-46. Published 2013 Aug 7. doi:10.1007/978-3-642-39374-7_1
5. India Pharmacopeia I Volume I, Government of India, Ministry of Health and Family welfare, Indian Pharmacopeia commission, 2014.
6. 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

7. Lukasz Komsta, Monika Waksmundzka-Hajnos, Joseph Sherma. Thin Layer Chromatography in Drug Analysis. CRC Press, Taylor and Francis.
8. Wagner H. Plant Drug Analysis. A thin Layer chromatography Atlas. 2nd ed. Heidelberg: Springer-Verlag Belgium; 2002:305, 227.

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