



Physicochemical Analysis in Standardization of Siddha Polyherbal Formulation *Sangamver Thailam*

Smilin Sherry V G^{1*}, Suresh K², Meenakshi Sundaram M³, Meenakumari R⁴

¹PG Scholar, Department of Kuzhanthai Maruthuvam, National Institute of Siddha, Chennai-600 047, India.

²Associate Professor, Department of Kuzhanthai Maruthuvam, National Institute of Siddha, Chennai-600 047, India.

³Professor, The Head of the Department, Department of Kuzhanthai Maruthuvam, National Institute of Siddha, Chennai-600 047, India.

⁴Director, National Institute of Siddha, Chennai-600 047, India.

*Corresponding author's E-mail: smilinsberry96@gmail.com

Received: 18-05-2024; Revised: 26-08-2024; Accepted: 04-09-2024; Published on: 15-09-2024.

ABSTRACT

This article presents a comprehensive physicochemical analysis of *Sangamver Thailam*, a Siddha polyherbal formulation used in traditional medicine to treat various diseases. The medicine was prepared as per the description mentioned in Siddha literature. The aim of the study is to ensure the nature and quality of study drug. The study was carried out based on PLIM guidelines. The formulation was subjected to various physicochemical evaluations including iodine value, saponification value, viscosity, refractive index, weight per ml, acid value, and peroxide value. Thin layer chromatography (TLC) and high-performance thin layer chromatography (HPTLC) analyses were also performed to investigate the presence of phytoconstituents. The findings reveal significant insights into the physical characteristics, solubility, and chemical stability of *Sangamver Thailam*, contributing to quality control and therapeutic efficacy assessments. The medicine is a yellowish, viscous liquid with greasy consistency and is soluble in chloroform and ethyl acetate. HPTLC results shows the presence of 8 distinct phytoconstituents, thus confirming the presence of multiple bioactive components in it. This baseline study ensures the quality of the medicine, based on which the future studies can be undertaken.

Keywords: Sangamver Thailam, Siddha Medicine, Physicochemical Analysis, Thin Layer Chromatography (TLC), High-Performance Thin Layer Chromatography (HPTLC), Herbal Formulations, Standardization.

INTRODUCTION

Siddha system of medicine, a traditional healing system originating from South India, encompasses a vast number of treatments for approximately 4448 types of diseases. Within the Siddha system, Thailam represents one of the 32 external medicines widely used for its therapeutic properties.¹ The medicine *Sangamver Thailam* is mentioned in ancient Tamil literature, specifically in the *Aathmaratchamirtham enum vaithya sarasangiragam*, for treating conditions like *Kuttam*, *Sori*, *Sirangu*, *Kiranthi*, *Soolai*, *Senkarappan*, *Vandukadi*, *Silavisham*, *Kshayam*, and *Irumal*.²

The medicine is made up of six herbals with *Sangamver* (*Azima tetracantha*) being the main ingredient. The rawdrugs were purified as per the procedure given in Siddha texts of *Sigicha ratna deepam*.³ All the drugs have potential health benefits and most of them are anti-oxidants. Siddha medicine has a long history of treating Skin diseases which are commonly described as *Kuttam*. Although the medicine has been indicated for various diseases, majority of indications are mentioned for Skin diseases. The shelf-life of the *thailam* form of medicine is one year.¹ Despite the general belief that all herbals are safe, there could be undesirable side effects.⁴ Though traditional formulations have a long history of use, modern evaluations are essential to ensure their safety, quality, and efficacy in contemporary settings. Standardization is defined as "The body of information and control necessary

to produce material of reasonable consistency".⁵ In this study the medicine is standardized as per PLIM guidelines using suitable analytical techniques.^{6,7} This study provides essential data on the formulation's stability and consistency, contributing to its validation as a safe and effective remedy in modern healthcare practices. The aim of this study is to identify the quality of *Sangamver Thailam*. The physical and chemical characters have been recorded. The solubility profile with various solvents have been done.⁸ To identify the phytochemical constituent techniques like TLC and HPTLC was done.^{9,10} Acid value and peroxide value was determined to know about the rancidity and oxidation of the medicine.

MATERIALS AND METHODS

Preparation

Ingredients:

Ingredients of *Sangamver Thailam* are provided in Table 1. All the raw drugs were purified as per the procedure given in the siddha literature namely *Sigicha Ratna Deepam*. *Sangamver* was ground as coarse powder in stone mortar and added to water (21.5L) and made into decoction of 1/8th part. Then gingelly oil was added to it. The remaining raw drugs were ground with cow's milk and added to the decoction-oil content and boiled until it reaches the oil consistency. The oil was filtered and cooled, then stored in an airtight container.



Table 1: Ingredients of *Sangamver Thailam*

Sl.no.	Tamil Vernacular Name	Botanical Name	Quantity
1.	<i>Sangamver</i>	<i>Azima tetracantha</i>	1/4 Thulaam (875 g)
2.	<i>Nallennai</i>	Gingelly oil	1 Padi (1.3L)
3.	<i>Milagu</i>	<i>Piper nigrum</i>	1 Palam (35 g)
4.	<i>Karunjeeragam</i>	<i>Nigella sativa</i>	1 Palam (35 g)
5.	<i>Elam</i>	<i>Elaterria cardamomum</i>	1 Palam (35 g)
6.	<i>Masikkai</i>	<i>Quercus infectoria</i>	1 Palam (35 g)
7.	<i>Pasum paal</i>	Cow's milk	As required

PHYSICOCHEMICAL EVALUATION

Physico chemical characters were evaluated as follows:

Organoleptic Characters

Characters like state, nature, odor, consistency, flow property and appearance of the study drug were noted.

Solubility Profile:

The study drug was mixed with different solvents to check the solubility profile.

Determination of Iodine value

About 20 gm weight equivalent of the 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. About 10 ml of KI solution was added to this and titrated against 0.1 N Sodium thiosulphate solutions until the appearance of yellow color. 1 ml of starch indicator was added and again titrated against the sodium thiosulphate solution from the burette. Disappearance of the blue color indicates the end point. The above procedure was repeated without taking a sample and the corresponding reading for blank titration is noted.

Determination of saponification value

About 2 gm weight equivalent 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. The same procedure was repeated without taking the sample for blank titration. Both the sample and blank round bottomed flasks were refluxed for 1 hour. After reflux, both the round bottomed flasks were allowed to cool. The samples were titrated using 0.5 N HCl with phenolphthalein indicator. The disappearance of pink indicates the end point.

Determination of Viscosity value

Viscosity determination was 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 below the upper reservoir, is measured.

Determination of Refractive Index

Determination of RL was carried out using Refractometer.

Determination of Weight per ml

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

Acid Value

Accurately 5 g weight equivalent of the 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 the 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 color indicated the end point. The volume of consumed KOH solution was determined and the titration of the 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 = Titer Value X 0.00561X 1000 / Weight of test sample (g)

Peroxide value

5 g weight equivalent of the substance was examined, accurately weighed, into a 250-ml glass-stoppered conical flask, then 30 ml of a mixture of 3 volumes of glacial acetic acid and 2 volumes of chloroform was added and swirled until it dissolved and 0.5ml volumes of saturated potassium iodide solution was added. It was allowed to stand for exactly 1 minute, with occasional shaking, then 30 ml of water was added and titrated gradually, with continuous and vigorous shaking, with 0.01M sodium thiosulphate until the yellow color almost disappears. 0.5 ml of starch solution



was added and the titration was continued then shaken vigorously until the blue color just disappears (a ml). The procedure was repeated omitting the substance being examined (b ml). The volume of 0.01M sodium thiosulphate in the blank determination must not exceed 0.1 ml.

$$\text{Peroxide value} = 10 (a-b) / w$$

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-microliter 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.

High Performance Thin Layer Chromatography Analysis:

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 analyzed. 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 fingerprint was developed for the detection of phytoconstituents present in each sample and their respective Rf values were tabulated.

OBSERVATION AND RESULTS

The results are tabulated in the following tables and figures.



Figure 1: TLC Visualization of SVT at 366 nm

Table 2: Physico Chemical parameters of *Sangamver Thailam*

Sl. No.	Parameters	<i>Sangamver Thailam</i>
1.	State	Liquid
2.	Nature	Viscous
3.	Odor	Mild
4.	Touch / Consistency	Greasy
5.	Flow Property	Free Flowing
6.	Appearance	Yellowish
7.	Viscosity at 50°C (Pa s)	61.92
8.	Refractive index	1.84
9.	Weight per ml (gm/ml)	0.73
10.	Iodine value (mg I ₂ /g)	120.65
11.	Saponification Value (mg of KOH to saponify 1 gm of fat)	184.68
12.	Acid Value mg KOH/g	1.009
13.	Peroxidase Value mEq/kg	4.86

Table 3: Solubility Profile

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

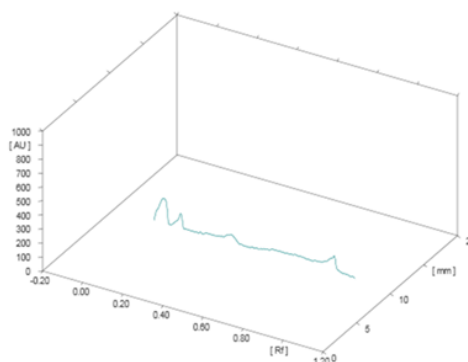


Chart 1: 3D – Chromatogram

Table 4: Peak Table

Peak	Start Rf	Start Height	Max Rf	Max Height	Max %	End Rf	End Height	Area	Area %
1	0.01	81.5	0.05	176.5	31.32	0.07	5.1	3042.7	27.39
2	0.08	2.7	0.13	102.3	18.15	0.18	0.3	1115.1	10.04
3	0.19	0.2	0.23	10.4	1.84	0.24	1.9	52.8	0.48
4	0.25	7.5	0.26	18.5	3.29	0.29	13.9	265.9	2.39
5	0.30	14.7	0.38	56.2	9.97	0.42	3.9	1548.6	13.94
6	0.52	11.8	0.59	37.7	6.69	0.61	36.0	921.1	8.29
7	0.63	35.4	0.70	45.6	8.08	0.72	39.4	1405.6	12.65
8	0.82	42.3	0.90	116.4	20.66	0.93	13.9	2757.3	24.82

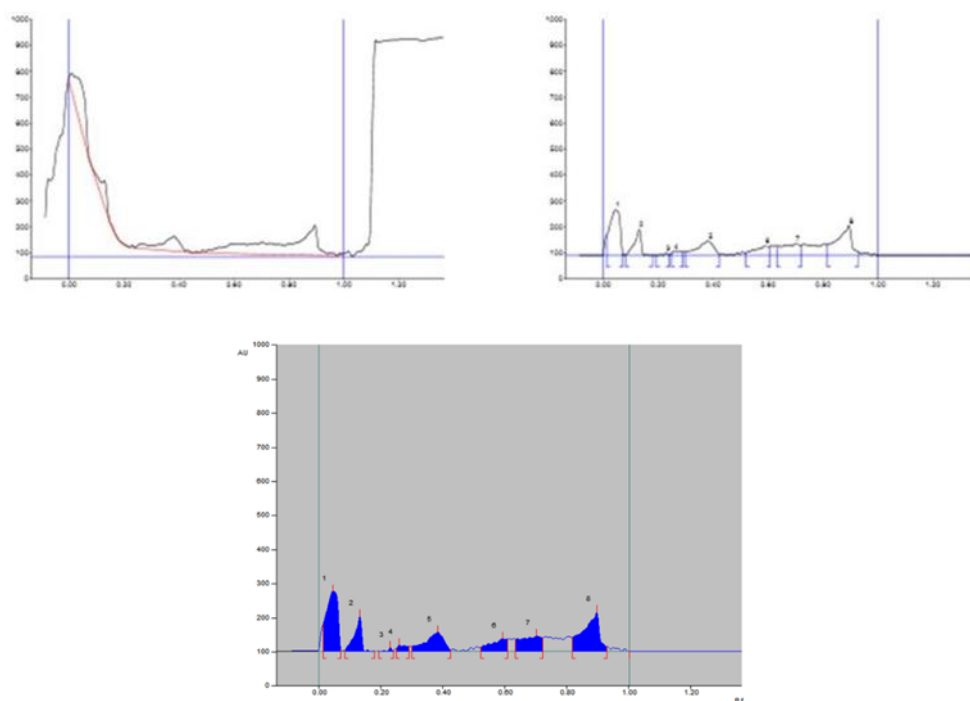


Chart 2: HPTLC fingerprinting of SVT

Interpretation

Organoleptic Characters:

The study shows that *Sangamver Thailam* is a yellowish liquid with mild odor, greasy consistency, yellowish in colour with free-flowing property.

Solubility profile:

The solubility profile shows that it is soluble in chloroform and Ethyl acetate. It is insoluble in Ethanol, Water, DMSO.

Physico chemical parameters:

The viscosity is 61.92, refractive index is 1.84, iodine value is 120.65, saponification value is 184.68, acid value is 1.009 and peroxidase value is 4.86 respectively.

TLC and HPTLC

TLC analysis was conducted using silica gel and different solvent systems. The sample was visualized under visible light and UV light at both 254 nm and 365 nm.

HPTLC analysis revealed the presence of eight prominent peaks, corresponding to the presence of eight distinct phytoconstituents. The R_f values (Retention factor) of the peaks ranged from 0.01 to 0.82, confirming the presence of multiple bioactive components in the formulation.

DISCUSSION

The physicochemical parameters obtained in this study confirms that *Sangamver Thailam* exhibits characteristics typical of a stable herbal oil-based formulation. The organoleptic characters suggests that it is a yellowish, viscous liquid that is greasy in consistency. The solubility profile shows that it is soluble in chloroform and ethyl

acetate. The iodine value reflects its unsaturation level, while the saponification value indicates its fatty acid content and potential for soap formation. The acid value and peroxide value suggest a relatively low degree of rancidity and oxidation, indicating the formulation's chemical stability under the given conditions. HPTLC fingerprinting analysis of the sample reveals the presence of eight prominent peaks corresponds to the presence of eight components present within it. R_f value of the peak ranges from 0.01 to 0.82. The chromatographic analysis, particularly the HPTLC fingerprinting, provided critical insights into the chemical complexity and possible therapeutic constituents of *Sangamver Thailam*, ensuring that it adheres to quality standards necessary for safe use.

CONCLUSION

The physicochemical analysis of *Sangamver Thailam* demonstrates that it is a chemically stable, well-characterized herbal formulation, exhibiting properties typical of traditional Siddha oils. The formulation has been evaluated for key quality indicators such as viscosity, refractive index, iodine value, saponification value, acid value, and peroxide value, all of which fall within acceptable limits for such preparations. The HPTLC fingerprinting analysis further validates the presence of multiple bioactive compounds, reflecting the formulation's potential therapeutic efficacy. This study emphasizes the importance of physicochemical evaluation in standardizing traditional medicines, ensuring their safety, quality, and effectiveness in modern therapeutic applications. Further studies involving biological assays are recommended to complement the physicochemical data and confirm the medicinal potential of *Sangamver Thailam*. With these

results the medicine may be subjected to in-vivo and in-vitro toxicity studies later.

Source of Support: The author(s) received no financial support for the research, authorship, and/or publication of this article

Conflict of Interest: The author(s) declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

REFERENCES

1. Thiagarajan R; Gunapadam Thathu – Jeeva Vaguppu; Vol.1, 1st edition; Chennai: Indian Medicine and Homeopathy Department; 2009;56-57.
2. Kanthasamy Mudaliar; Aathmaratchamirtham ennum vaithya saara sangiragam; first edition; Madurai; Sri Shenbaga publication; 2011;588-92.
3. Kannusamy pillai, Sigicha Rathana Deepam, second edition, Chennai, Rathna naicker and sons, 2018;20-35.
4. Kunle, Oluyemisi Folashade, Egharevba, Henry Omoregie and Ahmadu, Peter Ochogu; Standardization of herbal medicines- A review; international journal of Biodiversity and conservation, 2012;4(3):101-112, DOI: 10.5897/IJBC11.163
5. Shaukat Khalid, Kaneez Fatima, Hina Yasin, Rehana Perveen, Hina Abrar, Iqbal Ahmad; Standardization of herbal formulations: an overview; Baqai.J. Health science, 2014;17:1-2.
6. India Pharmacopeia, Volume I, Government of India, Ministry of Health and Family welfare, Indian Pharmacopeia commission, 2014.
7. 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. 2020.
8. Indian standard methods of sampling and test for oils and fats Indian standard institution New Delhi, 47-50. 1964
9. Lukasz Komsta, Monika Waksmundzka-Hajnos, Joseph Sherma. Thin Layer Chromatography in Drug Analysis. CRC Press, Taylor and Francis. 2022.
10. Wagner H. Plant Drug Analysis. A thin Layer chromatography Atlas. 2nd ed. Heidelberg: Springer-Verlag Belgium; 2002:305,227-32.

For any questions related to this article, please reach us at: globalresearchonline@rediffmail.com

New manuscripts for publication can be submitted at: submit@globalresearchonline.net and submit_ijpsrr@rediffmail.com

