

# STANDARDIZATION OF HERBO – MINERAL FORMULATION SOODHA VALLATHI URUNDAI THROUGH PHYSICO CHEMICAL ANALYSIS, PHYTO CHEMICAL ANALYSIS AND BIO CHEMICAL ANALYSIS.

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**Abstract-** Siddha medicine is an ancient form of medicine. In Siddha system of medicine, there are different types of medicines mentioned to treat various diseases. Among those different types, Soodha Vallathi Urundai is an internal medicine mentioned in the siddha literature *Agathiyar vaithiya vallathi – 600* which plays a special role in the treatment of such a cruel disease named Cancer. Nowadays cancer affects all age group and all genders. In medical science nowadays various medicines and chemotherapy treatment are appreciated to deal the cancer. But chemotherapy patients facing a lot of complications and side effects. Soodha Vallathi Urundai is a heavenly elixir provided by Siddhars to handle Cancer patients. Physio chemical, Phyto chemical, Organoleptic and Biochemical analysis and HPTLC for Sooda Vallathi Urundai was carried out according to PLIM guidelines and the values of Loss on drying 1.66%, Total Ash value - 2.92%, Acid insoluble Ash - 0.97%, along with HPTLC, Specific Pathogen, Sterility test results are also attached herewith. And in the future, toxicology studies and pharmacological studies can be done for Soodha Vallathi Urundai to treat cancer.

**Key Words :** Siddha medicine, Soodha Vallathi Urundai, Chemotherapy, HPTLC, PLIM.

## INTRODUCTION:

Cancer is known as *Puttru* in *Siddha* medicine. In this present world, people are suffering from various diseases. Among them, cancer is one of the cruel diseases. The current medical science is developing very fast and various diseases are being solved by it. Although some cancers are cured in the early stage, sometimes there are fatal. Chemotherapy is one of the best treatments for this, but their complications are remarkable. According to 2018 WHO study, in worldwide cancer is considered as the second fatal disease. It states, that 9.6 million people died because of cancer <sup>[1]</sup>. In India alone in 2022, 14,61,427 people were affected and 1 in 9 people were affected by lung cancer <sup>[2]</sup>. According to the year 2020, Oral cancer ranked 13th in the world, in which 3,77,713 new patients and 1,77,757 people died so far <sup>[3]</sup>. By the year 2020, Cervical cancer is the 4th most common cancer in the world with 6,04,000 new cases and 3,42,000 deaths <sup>[4]</sup>. In Siddha medicine, cancer and its pathophysiology, treatment methods are discussed. In Siddha medicine, Mercury-based drugs are often used for cancer. *Soodha Vallathi Urundai* a herbo-mineral formulation, which is indicated for cancer, mentioned in Siddha literature *Agathiyar Vaidya Vallathi 600*.

## MATERIALS AND METHODS

### Ingredients of the drug

Table No : 01

S.NO	NAME OF THE DRUG	SCIENTIFIC NAME	QUANTITY
1.	Serangkottai	<i>Semicarpus anacardium</i>	24 varagan ( 98.4 g )
2.	Thaneervittan kizhangu	<i>Asparagus racemosus</i>	2 varagan ( 8.4g )
3.	Kurasani Omam	<i>Hyoscyamus niger</i>	2 varagan ( 8.4g )
4.	Nilapanai kizhangu	<i>Curculigo orchioides</i>	2 varagan ( 8.4g )
5.	Rasam	<i>Hydragyrum</i>	2 varagan ( 8.4g )
6.	Pooram	<i>Hydragyrum subchloride</i>	2 varagan ( 8.4g )

### Drug selection:

*Soodha vallathi urundai* is given in the book *Agathiyar Vaidya Valladi - 600* as an excellent medicine for cancer.

### Collection of the Drugs :

All Raw Drug products given above are bought from Authenticated Naattu marunthu kadai Parry's, Chennai.

### Authentication of Drugs:

All medicinal ingredients in Soodha Vallathi Urundai have been authenticated by Gunapadam experts and botanist and the labeled No is given here.

**Purification of the drug:**

Purification process were done as per classical Siddha Literature SARAkkUGALIN SUTHTHI MURAI.

**Purification of Mercury:**

35g of mercury hydrargyrum is pounded with brick dust and turmeric powder for an hour and cleansed with purified water. Then it was boiled with *Acalypha indica* leaf extract. until the water content of the extract evaporates [5].

**Purification of hydrargyrum sub chloride:**

8.75 grams of each betel leaf and pepper taken, minced with water and made into a fine paste and mixed with 1.3 l of water. The Hydrargyrum subchloride which have to be purified is wrapped in a cotton cloth and immersed into the mixture and burnt in low flame until the water evaporate to three fourth of its original quantity used. Then the subchloride is taken, wiped out and air dried [6].

**Purification of Semecarpus anacardium :**

If Serangottai milk gets on our body, we will get blisters. Therefore, if the milk gets on our body, we should cut off the part like nose and throw it away and put it in milk and boil it and wash it. [7]

**Preparation of Soodha Vallathi Urundai:**

2 varagan (8.4g) of each Purified *Kurasani omam*, *Thannirvittan kizhangu*, *Nilapanai kizhangu* are dried in the daylight and pulverised in an iron mortar for 48 minutes, followed by the addition of Purified *Serangottai* one by one to the mixture. Then 2 varagan (8.4g) Purified *Soodham* and *Pooram* are added to the grinded matter. The mixture is then taken along in the consistency of a fine paste and rolled up in the form of tablets of 22 nos and named as SVU [8].

**Storage of the drug:**

The prepared test drug was stored in a clean, dried, air tight container. The contents were explored frequently to avoid moisture and microbes.

**ADMINISTRATION OF DRUG:**

**Form of the medicine:** Tablet

**Route of Administration:** Enteral route

**Dose:** 1 pill (2gm) three times for 7 days

**Indication: Puttru ( Cancer )**, Kuttam, Araiappu, Soolai, Pavuthram, Pilavai, Vippurithi, Kandamalai.

**Organoleptic Character:**

Color, Odor, Taste, Texture, Particle Size and Other Morphology

**Qualitative analysis of SVU :**

Qualitative analysis of SVU is done as per PLIM Guidelines. Namely Physio Chemical analysis, Phyto chemical analysis, Biochemical analysis, Sterility test, High Thin Layer Chromatography, Pesticide residue test, Heavy metal analysis, Aflatoxins etc. studies have been completed at Noble research institute, Perambur, Chennai.

**Physio chemical analysis : [9 – 12]**

Physio chemical analyzes of SVU such as Loss on drying, Total ash, Acid insoluble Ash, Water soluble ash, Alcohol soluble extractive and Water soluble extractive have been evaluated<sup>[9]</sup>. And pH have been evaluated<sup>[10]</sup>, also studies like Uniformity weight have been done<sup>[11]</sup>. Along with Solubility test also done here<sup>[12]</sup>.

**Phyto chemical analysis:**

Phyto chemical analysis by PLIM guidelines through which Alkaloids, Tannins, Flavonoids, Saponins, Glycosides, Coumarins, Phenols, carbohydrates, Cyanins, Steroids, and triterpenoids have been detected [13].

**TLC and HPTLC analysis:**

The quality of SVU is assessed by TLC (Thin Layer Chromatography) and HPTLC (High Thin Layer Chromatography) [14-15].

**Heavy metal analysis by Atomic Absorption**

Spectrometry method (AAS): Mercury, Cadmium Lead and Arsenic have been detected by AAS method [16-17].

**Bio Chemical analysis of Basic radicals and Acidic Radicals:**

For evaluated of Carbonates, Zinc, Copper, Phosphates, Sulfides and Chloride [18].

**Sterility test:**

Organisms in SVU are detected by the pour plate method [19].

**Specific Pathogens Test :**

EMB agar - *E. coli*

Deoxychelate agar - *Salmonella*

Mannitol Salt agar - *Staphylococcus Aureus*

Cetrimide agar - *Pseudomonas Aeruginosa*

It has been studied whether the above pathogens are present through special agar [20].

**Pesticide residue Analysis : [21 - 22]**

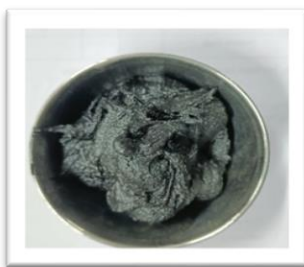
Pesticide residues such as Organo Chlorine Pesticides, organo phosphorus pesticides, organo carbamates, pyrethroid were tested.

**Aflatoxin analysis:**

Aflatoxins like B1, B2, G1, G2 were detected by TLC analysis [23].

**RESULTS :**

A drug Soodha Vallathi Urundai is a tablets are dark black in colour, shiny appearance, characteristic odour, non sticky nature are present. There were noted in Tab No : 02.



**Fig No : 01** Prepared form of SVU

**Tab No : 02 Results of organoleptic characteres**

S.NO	VARIABLE	RESULTS
01.	Color	Dark black
02.	Odor	Characteristic
03.	Taste	Bitter
04.	Touch	Soft
05.	Nature	Non – sticky
06.	Appearance	Shiny

**Tab No : 03 Physio chemical analysis of SVU**

S.NO	PARAMETERS	PERCENTAGE
01.	Loss on drying	1.66%
02.	Total ash value	2.92%
03.	Acid insoluble ash	0.97%
04.	Water soluble ash	28.26%
05.	Water soluble extraction	6.23%
06.	Alcohol soluble extraction	16.71%
07	Ph	6.5
08.	Particle size	154.5± 20.16µm

**Tab No : 04 Average weight of sample SVU**

Table No	Tablet Weight	Percentage Deviation
1	0.364	0.91
2	0.436	14.81
3	0.334	-4.88
4	0.322	-7.20
5	0.293	-12.80
6	0.347	-2.37
7	0.407	9.21
8	0.378	3.61
9	0.35	-1.80
10	0.327	-6.24
11	0.408	9.40
12	0.394	6.70
13	0.367	1.49
14	0.328	-6.04
15	0.397	7.28
16	0.375	3.03
17	0.339	-3.92

18	0.369	1.87
19	0.321	-7.39
20	0.33	-5.66
<b>Average</b>	<b>0.3593</b>	

**Tab No : 05 Percentage Deviation on uniformity of Weight of the sample SVU**

Average Weight of Tablet/ urundai	Number of Tablets/ urundai	Deviation (%)	Pharmacopoeial Category	Limit
0.3593 mg	4	0 – 3 %	250 mg and more	± 5% for Minimum 18 Tabs ± 10% for Maximum 02 Tabs
	6	Above 3 %		
	10	Negative deviation value -1.79 to -12.79		

**Solubility Analysis of SVU :****Tab No : 06 Solubility Profile**

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

**Phyto chemical analysis :**

Alkaloids, Carbohydrates, saponins and Gum & Mucilage in SVU Formulation have been found through Qualitative Phyto chemical analysis.

**Tab No : 07**

S.NO	PHYTO CHEMICALS	H <sub>2</sub> O EXTRACT
01.	Alkaloids	+ve
02.	Carbohydrates	+ve
03.	Saponin	+ve
04.	Gum & mucilage	+ve

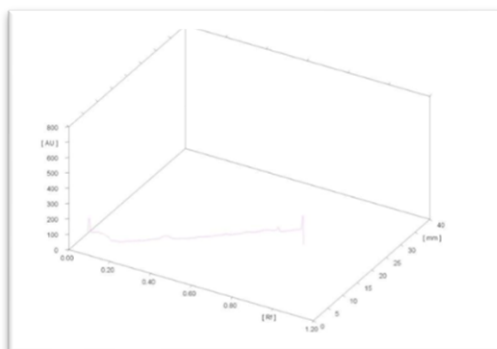
**Phyto chemical analysis****Fig No : 02**

**TLC and HPTLC analysis:**

High Thin Layer Chromatography analysis has evaluated 5 eminent peaks which corresponds to the presence of 5 versatile phytochemical components.

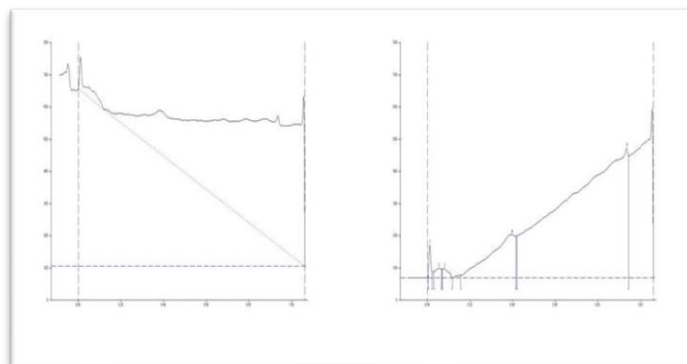
**TLC Visualization of SVU at 366 nm**

**Fig No : 03**  
**3D - Chromatogram**



**Fig No : 04 HPTLC finger printing of Sample SVU**

**Fig No : 05**



Tab No : 08 Peak Table

Peak	Start Rf	Start Height	Max Rf	Max Height	Max %	End Rf	End Height	Area	Area %
1	0.00	10.9	0.01	102.4	14.62	0.02	16.8	579.7	0.73
2	0.03	20.4	0.05	30.2	4.31	0.07	27.7	504.0	0.64
3	0.07	27.8	0.08	29.9	4.27	0.12	1.1	501.1	0.63
4	0.15	7.8	0.40	134.8	19.24	0.42	130.0	9383.6	11.88
5	0.42	130.3	0.94	403.4	57.56	0.95	379.2	68024.0	86.11

**Biochemical analysis :**

Through this study, it has been found that Acid radicals, Basic radicals in SVU are respectively Carbonates, Phosphate, Lead and Mercury.

Tab No : 09 Analytical Investigation on Test for Acid Radicals

S.NO	TEST	INFERENCE	RESULTS
1.	Carbonates	Creation of brisk effervescence	Present
2.	Phosphates	Creation of yellow precipitate	Present

Tab No : 10 Analytical Investigation on Test for Basic Radicals

S.NO	TEST	INFERENCE	RESULTS
1.	Mercury	Presence of yellow precipitate	Present
2.	Copper	Presence of blue precipitate	Present

Tab No : 11 Heavy metal analysis

Name of the HeavyMetal	Absorption Max $\lambda$ 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	0.25 PPM	1 ppm

Below Detection Level

**Sterility Test :**

Sterility Testing pour plate method shows that there is no growth or colonies from the inoculated plate.

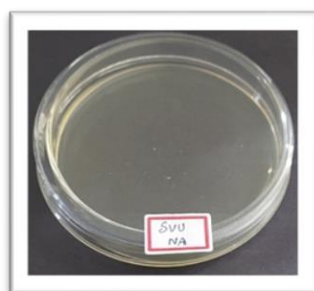


Fig No : 06

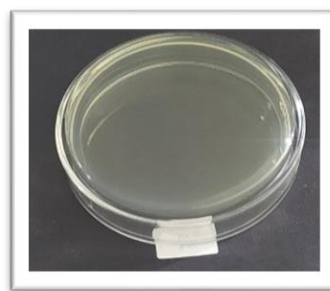


Fig No : 07

**Tab No : 12 Sterility Test**

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

**Specific pathogen testing:**

When SVU is subjected to this test, it is seen that there is no growth and no colonies when looking at the inoculated plate.

**Tab No : 13 Specific Pathogen Analysis**

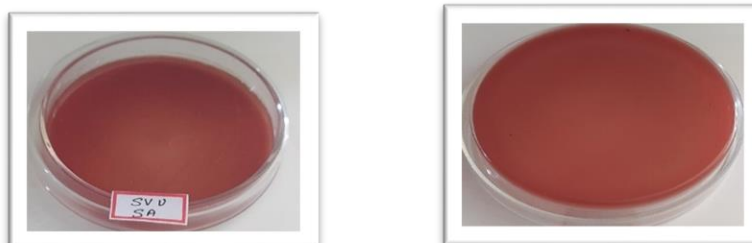
Organism	Specification	Result	Method
<i>E-coli</i>	Absent	Absent	As per AYUSH specification
<i>Salmonella</i>	Absent	Absent	
<i>Staphylococcus Aureus</i>	Absent	Absent	
<i>Pseudomonas Aeruginosa</i>	Absent	Absent	

*Culture plate with E-coli (EC) specific medium*



**Fig No : 09**

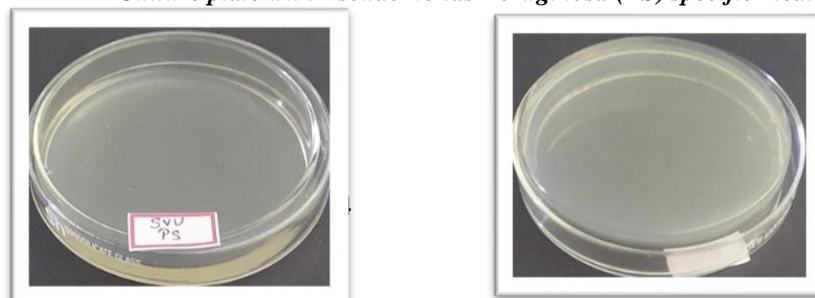
*Culture plate with Salmonella (SA) specific medium*



*Culture plate with Staphylococcus Aureus (ST) specific medium*



*Culture plate with Pseudomonas Aeruginosa (PS) specific medium*



**Pesticide Residue Analysis:**

Through this study very low amount of pesticide residue is found in SVU.

**Tab No : 14 Test Result Analysis of the Sample SVU**

Pesticide Residue	Sample SVU	AYUSH Limit (mg/kg)
<b>I.Organo Chlorine Pesticides</b>		
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
<b>II.Organo Phosphorus Pesticides</b>		
Malathion	BQL	1mg/kg
Chlorpyriphos	BQL	0.2 mg/kg
Dichlorovos	BQL	1mg/kg
<b>III. Organo carbamates</b>		
Carbofuran	BQL	0.1mg/kg
<b>III.Pyrethroid</b>		
Cypermethrin	BQL	1mg/kg

BQL- Below Quantification Limit.

**Aflatoxin Testing :**

Aflatoxin analysis by TLC plate analysis showed very low levels of Aflatoxin B1, B2 and Aflatoxin G1, G2 as per PLIM guidelines.

**Tab No : 15 Aflatoxin Analysis**

Aflatoxin	Sample SVU	AYUSH Specification Limit
B1	Not Detected – Absent	0.5 ppm (0.5mg/kg)
B2	Not Detected – Absent	0.1 ppm (0.1mg/kg)
G1	Not Detected – Absent	0.5 ppm (0.5mg/kg)
G2	Not Detected – Absent	0.1 ppm (0.1mg/kg)

**Discussion :**

SVU is an excellent tablet prepared based on Siddha medicine method. It has the qualities of dark black in color, shiny appearance, characteristic odor, non-sticky and soft in nature. Also it has a Loss on drying of 1.66%, so its shelf life is long. Since Total Ash value is 2.92%, it shows that there are no impurities in it. Acid insoluble Ash is 0.97% and Water insoluble Ash is 18.26%, so Siliceous matter is very less. Water soluble extraction and Alcohol soluble extraction are 6.23% and 16.7% respectively, so SVU is of good quality. Based on the evaluation, the uniformity of weight was determined by using SVU urundai in order to make sure that the dosage units is consistent. From the result obtained, it was observed that the tablets used has an average weight of 0.3593 gm which is under the category of '250 mg and more' as per pharmacopeia limit. Alkaloids play a major role in cancer medicine.



Mainly alkaloids are used in cancer medicine because of their antiproliferative and anti-cancer effects<sup>[24]</sup>. Saponins are slightly more complex and different in their structure. It induces apoptosis and inhibits the growth of cancer cells, acts as Anticancer compound in cell line<sup>[25]</sup>. Gum and Mucilage is a material of the plant. They are all good for burns, wounds, sores, especially external and internal inflammations and irritation<sup>[26]</sup>. Copper-containing drugs are used as monotherapy and in combination with other drugs for cancer chemotherapy and exert inhibitory action on topoisomerases involved in the regulation of DNA Topology<sup>[27]</sup>. Through the sterility test, it is found that there are no microorganisms and through the pour plate method, there are no specific pathogens such as *E. coli*, *Salmonella*, *Staphylococcus Aureus* and *Pseudomonas Aeruginosa*. Pesticide residue analysis does not contain any molecules such as Organochloride, Organophosphorus and Organocarbonates which are harmful to life. Through Aflatoxin analysis, it has been found that SVU does not contain any Aflatoxin spores from the genus *Aspergillus*.

#### Conclusion :

In Siddha medicine, various medicines are given for cancer. Among them, Soodha Vallathi Urundai is given for cancer. For this, Physiochemical studies like Loss on drying, Total ash value, Water soluble ash, Alcohol soluble ash, Water soluble extraction, Alcohol soluble extraction etc. and Phyto chemical studies like Alkaloids, Carbohydrates, Saponins, and Gum and Mucilage have been identified and standardized. Although Alkaloids and Saponins have the ability to cure cancer, in the future studies such as Cell line study, Acute toxicity and Oral toxicity will be done to provide better treatment for cancer.

#### Declaration by Authors :

##### Ethical approval

Approved

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##### Conflict of Interest

No conflict of interest

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