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Research Article

PHARMACEUTICO-ANALYTICAL STUDY OF *VISHATINDUKA TAILA*

Vanitha V[1]*, B. Saravanan[2], K.L. Virupaksha Gupta[3]

*[1]PG Scholar, [2]Associate Professor, [3]HOD & Professor, Department of Rasashastra and Bhaishajya Kalpana, Sri Jayendra Saraswathi Ayurveda College and Hospital, Chennai-Bangalore Highway, Nazarathpet, Chennai, Tamilnadu, India.

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ABSTRACT

Vishatinduka Tailam is mentioned in context of *Vataroga nidhanalakshana chikitshaadhyana* of *Basavarajiyam* text for the treatment of *Sarvangasandhiasthigatavata, Amlavata, Sula, Anulomanavayu, Sthambha, Dhanurvata, Kaphajavikaras*, severe painful condition of the body, etc., It is prepared with ingredients like *Vishatinduka, Jambira, Aranala, Tilataila* and *Erandataila.* It is an attempt made to validate the pharmaceutical and analytical parameters of *Vishatinduka taila*. Three batches of *Vishatindukataila* were prepared. Pharmaceutical study of batches and its standardization was done. It took five days for preparation of each batch due to presence of *Aranala*. Intermittent cooling was done. At the end of *Taila* preparation, mustard brown coloured oil was obtained and strong odour present. All *Sneha siddhi lakhanas* were observed and the loss was approximately 10% in all the three batches of *Taila*. To establish standards of *Vishatinduka Taila,* physicochemical and chromatographical methods were performed. The results found are acid value 12.344mg/KOH/g, peroxide value 5Meq/kg, Density 0.932g/cm^3, Specific gravity 0.932, pH 4.5+/_0.3, Loss on drying 0%, Refractive index 73.5+/_0.2% brix, saponification value 112.22mg KOH/1g and Total fatty matter 95.4%. Physicochemical test was done in first and second month also results obtained. In HPTLC evaluation variable number of spots are visualized. It showed the presence of four phytoconstituents.

INTRODUCTION

Vishatinduka (*Strychnosnux-vomica* Linn.), commonly known as *Kuchala* belongs to the family Loganiaceae, is a medium-sized poisonous tree mentioned in various *Samhithas* of Ayurveda and is one amongst *Upavishas* mentioned in textbooks of *Rasashastra*.[1] It has properties of *Kaphavatahara, Visaghna, Grahi* etc. It has nerve tonic and stimulant properties. This action is due to alkaloid strychnine, which increases the reflex excitability of the spinal cord. *Strychnusnux-vomica* is considered to be specifically effective in spinal conditions.[2] External application of *Vishatinduka* in the form of *Taila* might be very effective in spine related disorders. So, *Taila* is prepared by using *Vishatinduka* and study conducted.

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Another method of preparation of *Vishatinduka Taila* was mentioned in Bhaishaijya Ratnavali reference.[3]

Vishatinduka Taila is oleaginous preparation mentioned in classics context of *Basavarajiyam* text and it is effective in management of *Vatavyadhi* like, *Vatarakta, Suptavata, Kushta, Vaivarnya, Sarvangavata, Sandhivata, Asthigatavata, Amlavayu, Kaphamaya, Anilaghorashoola, Vilomavayu, Anulomavayu, Sthambhavata* and *Dhanurvata*.[4]

In *Vishatinduka Taila*, *Kupilu* is the major ingredient. Therapeutic usage of poisonous drugs in various formulations is done after a particular pharmaceutical procedure called as *Shodhana.* Many Acharyas as described the efficacy of *Shodhana* of *Kupilu* in form of *Vati, Churna* etc. But till now the studies have not been conducted on *Vishatinduka Taila*. Pharmaceutical study of *Vishatinduka Taila* and its analytical parameters are studied in this study. By doing analytical study of this *Taila,* the organoleptic characteristics, physio-chemical and chromatographical parameters are studied.

MATERIALS AND METHODS

Pharmaceutical Study

Ingredients: 1. *Vishatindukab*ija (*Nux-vomica* seeds), 2. *Aranala* (sour gruel) 3. *Jambirarasa* (fresh lemon juice) 3. *TilaTaila* (sesame oil) 4. Eranda *Taila* (castor oil). [Figure 1]

Collection of Raw Materials: Strychnosnux-vomica seeds were collected at forest of Tirumala hills near Tirupathi. *Jambira*, *Tila Taila* and *Eranda Taila* were bought from the local market of Chennai. *Kupilu* was authenticated at Department of *Dravyaguna* and preparation of *Vishatinduka Tailamm* was carried out in *Rasashastra* and *Bhaishajyakalpana* department, Sri Jayendra Saraswathi Ayurveda college and hospital, Chennai.

***Vishatinduka Tailam* Preparation:** *Vishatinduka Tailam* preparation was done in three phases. They are a) preparation of *Aranala* b) *Shodhana* of *Vishatinduka* and c) Preparation of *Vishatinduka Tailam*. *Aranala* was prepared as per reference of Bhavaprakasha[5], *Vishatindukabija shodhana* was done as per the reference of *Rasa Tarangini*[6] and *Vishatinduka Tailam* was prepared.

a. Preparation of *Aranala*: Jowar (barley) was grinded to make coarse powder form and was immersed *(Nimajjana)* in a stainless-steel vessel containing water. It took 14 days for the fermentation process. After fermentation *Aranala* was filtered.

b. *Shodhana* of *Vishatindukabija*: *Vishatindukabija* were immersed in a stainless-steel vessel containing *Aranala.* Every day the *Aranala* was changed and replaced with the new one. The process was carried out for 3 days, later it was taken out and washed 3 times with hot water and the outer covering of the seeds are removed and dried under sunlight.

c. Preparation of *Vishatinduka Taila*

Apparatus: Pounding machine, gas stove, containers, tray, clean fine cloth, spatula.

Ingredients:

- *Vishatindukabeeja* – 800gm
- *Vishatindukabeejachoorna*– 800gm
- *Jambeeraswarasa* – 1600ml
- *Aranala* – 1600ml
- *Tila Taila* – 1600ml
- *Eranda Taila* – 800ml

Same quantity of ingredients was taken to prepare each batch.

Shodhita Vishatinduka beeja were taken and kept soaking in *Aranala* which is prepared by using jowar for a day. Often it is stirred well and *Aranala* was decanted and this was used in the preparation of this *Taila*. *Tila Taila* and *Eranda Taila* was added in *Tailapatra* and heated mildly. In this *Tailapatra*, *Aranala, Jambeeraswarasa, Vishatindukabeejachoorna,* were poured and *Tailapaka* was carried out until the *Snehasiddhi Lakshanas* of *Taila* are observed. Then it is filtered and stored. [Figure 2]

Table1: Observations during *VishatindukaTailam*

Day	Time	Observation 1st batch	Observation 2nd batch	Observation 3rd batch
1st day	1st hr	Oil was heated until appearance of fumes. Then allowed for self-cooling.	Oil was slightly heated about 30 mins and allowed for self-cooling.	Oil was slightly heated about 30 mins and allowed for self-cooling
	2nd hr	*Kalka* was added, brown colour *Kalka* floated over peripheral surface of oil.	*Swarasa, Aranala* and *Kalka* was added, brown colour *Kalka* floated over peripheral surface of oil.	*Swarasa, Aranala* and *Kalka* was added, brown colour *Kalka* floated over peripheral surface of oil.
	3rd hr	Typical *Kupilu* smell was appreciated.	Typical *Kupilu* smell was appreciated.	*Kupilu* smell was appreciated.
	4th hr	Oil started to boiling	Oil started to boiling	Oil started to boiling
	5th hr	Froth started to appears.	Froth started to appears.	Froth started to appears.
	6th hr	Vigorous boiling was started, vaporization become prominent.	Vigorous boiling was started, vaporization become prominent.	Vigorous boiling was started, vaporization become prominent.
	7th hr	Stirring was done.	Boiling continued, heating was stopped.	Boiling continued, heating was stopped.
	8th hr	Boiling continued, heating was stopped after 8 hours.	Heating was stopped after continuous stirring process.	Heating was stopped after continuous stirring.
2nd day	1st hr	*Kalka* completely soaked in oil; *Kalka* sticked to the sides	*Kalka* completely soaked in oil; *Kalka* sticked to the	*Kalka* completely soaked in oil; *Kalka* sticked to the sides

		of the vessel	sides of the vessel	of the vessel
	2nd hr	Brownish coloured oil	Brownish coloured oil	Brownish coloured oil
	3rd hr	Strong smell of *Kalka* was observed	Strong smell of *Kalka* was observed	Strong smell of *Kalka* was observed
	4th hr	Vaporization was increased	Vaporization was increased.	Vaporization was increased
	5th hr	Vigorous boiling started	Boiling started	Boiling started
	6th hr	Boiling was vigorous	Boiling was vigorous	Boiling was vigorous
	7th hr	Volume of liquid was reduced	Heating was stopped	Heating was stopped
	8th hr	Heating was stopped after 8 hours	Volume of liquid was reduced	Volume of liquid was reduced
3rd day	1st hr	Mustard brown colour of oil	Mustard brown colour of oil	Mustard brown colour of oil
	2nd hr	Bulk of *Kalka Dravya* is increased	Bulk of *Kalka dravya* is increased	Bulk of *Kalka* dravya is increased
	3rd hr	Boiling sound was more prominent	Boiling sound was more prominent	Boiling sound was more prominent
	4th hr	Brown colour with bubbles was observed	Brown colour with bubbles was observed	Brown colour with bubbles was observed
	5th hr	Vaporisation was increased	Vaporisation was increased	Vaporisation was increased
	6th hr	Continuous vaporisation process.	Continuous vaporisation process.	Strong smell of oil was observed
	7th hr	Strong smell of oil was observed	Strong smell of oil was observed	Strong smell of oil was increased
	8th hr	Heating was stopped	Heating was stopped	Heating was stopped
4th day	1st hr	Strong smell of oil was appreciated	Strong smell of was appreciated	With bubbles oil started to spill out
	2nd hr	Oil started to spill out with bubbles	Oil started to spill out with bubbles	Volume of liquid was completely reduced
	3rd hr	Volume of liquid was completely reduced	Volume of liquid was completely reduced	*Kalka* is moisture free
	4th hr	*Kalka* is moisture free	*Kalka* is moisture free	Oil started to spill out with bubbling
	5th hr	Continuous stirring was done to avoid sticking of *Kalka*	Continuous stirring was done to avoid sticking of *Kalka*	Continuous stirring was done to avoid sticking of *Kalka*
	6th hr	Gradual boiling was done	Gradual boiling was done	Temperature was checked
	7th hr	Temperature was checked	Temperature was checked	Gradual boiling was done
	8th hr	Heating was stopped	Heating was stopped	Heating was stopped
5th day	1st hr	No sound heard when *Kalka* was placed over the fire	No sound heard when *Kalka* was placed over the fire	No sound heard when *Kalka* was placed over the fire
	2nd hr	*Kalka* test was positive	*Kalka* test was positive	*Kalka* test was positive
	3rd hr	Froth appeared over the surface of oil	Froth appeared over the surface of oil	Over the surface of oil froth starts to appear.
	4th hr	Remarkable greenish brown colour of oil	Remarkable greenish brown colour of oil	Remarkable greenish brown colour of oil
	5th hr	Aroma was observed	Aroma was very strong	Aroma was observed
	6th hr	Aroma was strong	*Siddhi lakshana* of *Taila*	*Taila siddhi lakshana* attained

			observed	
	7th hr	*Taila siddhi lakshana* attained	Good aroma was observed	Good aroma observed
	8th hr	Intense heat was given	Intense heat was given	Intense heat was given
6th day		*Taila* was heated slightly, filtered and stored	*Taila* was heated, filtered and stored	*Taila* was heated, filtered and stored

Table 2: Results of *Taila* obtained

Batches	*Taila* obtained (ml)	*Taila* obtained (%)	Loss	Loss (%)
Batch 1	2150	89.58 %	250 ml	10.41%
Batch 2	2160	90%	240 ml	10%
Batch 3	2160	90.62%	225 ml	9.6%

Precautions

- The vessel used for the process should be clean and of adequate size, in order to avoid spilling of *Taila* because of excess foaming during *Paka.*
- *Mandagni* should be maintained throughout the process.
- Continuous stirring should be done to avoid the sticking of *Kalka* to the vessels.
- Timely performance of the *Paka Siddhi Parikshas* and observations of *Siddhi Lakshanas*
- Clean cloth should be taken for filtration purpose.
- *Taila* should be filtered immediately (warm condition) to avoid the loss. *Taila* should be filled in the suitable container.

Analytical study

Analytical assessment of *Vishatinduka Taila* was carried out to progress the basic standards. The *Taila* samples were analyzed first to develop the organoleptic standards. The organoleptic characters are tested with the help of sensory organs. These are colour, taste, odour, appearance, touch and clarity.

The colour was observed with the help of eye and odour was smell. Appearance, touch and clarity were observed by touching the *Taila*. The *Taila siddhi lakshanas* was also observed.

Physico-chemical parameters such as acid value[7], refractive index, saponification value, peroxide value[8] and HPTLC of *VishatindukaTailam* sample was done at laboratory of chimertech innovations, Chennai, Tamilnadu.

Table 3: Organoleptic characters of *VishatindukaTailam*

Physical test	*Vishatinduka Tailam*
Colour	Mustard brown
Taste	-
Odour	Characteristic strong
Texture	Viscous liquid
Touch	Unctuous
Clarity	Transparent

Table 4: Analytical parameter of *VishatindukaTailam*

Tests	**Results**	**After 1st month results**	**After 2nd month results**
Acid value	12.344mg/KOH/g	12.90mg KOH/g	12.90mg KOH/g
Peroxide value	5 mEq/kg	5 mEq/kg	5 mEq/kg
Density	0.932g/cm^3	0.932g/cm^3	0.930g/cm^3
Specific gravity	0.932	0.932	0.930
pH	4.5+/_0.3	4.5+/_0.3	4.5+/_0.3
Loss on drying	0%	0%	0%
Refractive index	73.5+/_0.2% brix	73.5+/_0.2%	73.5+/_0.1%
Saponification value	112.22mg KOH/1g	117.83mg KOH/1g	115.02mg KOH/1g

Total fatty matter	95.4%	95.4%	94.4%
Rancidity	Absent	Absent	Absent
Iodine value	99.48 gl.	99.48 gl.	101.52 gl.

HPTLC Analysis: The sample was dissolved in 1 ml suggested solvent and vortexed. This solution was used as test solution for HPTLC analysis. A 2ml aliquot of the above test solution were loaded as 5 mm band length in a 3 10 silica gel 60F254TLC plate using a Hamilton syringe and LINOMAT 5 instrument (CAMAG, Muttenz, Switzerland). The samples-loaded plate was kept in TLC twin through developing chamber (after saturation with solvent vapor) with respective mobile phases and the plate was developed in the respective mobile phase up to 90mm. The developed plate was dried by hot air to evaporate solvents from the plate. The plate was kept in a photo-documentation chamber (CAMAG REPROSTAR 3) and the images were captured in white light, and at 254mm. The developed plate was sprayed with respective spray reagent and dried at 100PC in a hot air oven. The plate was documented in daylight and at UV 366nm mode using the photo documentation chamber. After derivatization, the plate was fixed and scanning was done at 500 nm by TLC Scanner 3. The peak table, peak display and peak densitogram were recorded.

HPTLC Results: The HTPLC of *VishatindukaTailam* specific in the refractive domain done in n-butanol: acetic acid: water – 4: 1: 5 solvent system.

Derivative method used was UV254nm

Result: HPTLC finger printing showed that the presence of 4 phytoconstituents. [Figure 3]

Table 5: Interpretation for the HPTLC finger printing analysis

Peaks	Rf value
B1	0.37
B2	0.24
B3	0.11
B4	0.04

It has given the following results.

Peak 1– Start Rf was 0.00, start height was 23.9, maximum Rf was 0.01, maximum height was 185.8, maximum percentage was 11.96, end Rf was 0.03, end height was 0.0, area was 1154.1 and area percentage was 1.41.

Peak 2– Start Rf was 0.04, start height was 1.0, maximum Rf was 0.09, maximum height was 260.4, maximum percentage was 16.76, end Rf was 0.11, end height was 228.0, area was 7730.4 and area percentage was 9.44.

Peak 3– start Rf was 0.11, start height was 228.3, maximum Rf was 0.16, maximum height was 297.8, maximum percentage was 19.17, end Rf was 0.21, end height was 243.5, area was 18766.8 and area percentage was 22.92.

Peak 4– Start Rf was 0.24, start height was 240.7, maximum Rf was 0.34, maximum height was 425.1, maximum percentage was 27.36, end Rf was 0.37, end height was 362.8, area was 33540.5 and area percentage was 40.96.

Peak 5– Start Rf was 0.37, start height was 363.4, maximum Rf was 0.39, maximum height was 384.5, maximum percentage was 24.75, end Rf was 0.48, end height was 2.5, area was 20698.2 and area percentage was 25.28. [Figure4]

DISCUSSION

Sneha paka is the preparation of particular *Taila* with the help of *Kalkadravya, Dravadravya* and *Kashaya dravya* in specific ratio and heated at a particular temperature and duration till the completion of *Taila* preparation. The principle is transfer of active constituents of herbs in lipid and water according to its solubility.

For preparation of *Vishatinduka Tailam* in three batches, a wide mouthed *Tailapatra* was taken. *Tila Taila* and *Eranda Taila* added and preheating was done till fumes starts to appearing. This *Murchana* process avoids the *Amathva* of *Taila*. Microbial and fungal contamination also gets removed. Along with that moisture contamination and bad odour gets removed. Preheating was done for achieving above objectives. After cooling *Aranala* and *Jambiraswarasa* and *Kupilu Kalka* were added to *Murchita Taila* and heated.

Sneha Paka should not be finished in one day. It is told that basing on the *Dravadravya* duration of heating is mentioned in text *Bhaishajya Ratnavali.* As per it, *Sneha* which possess *Takra* or *Aranala* as *Dravadravya* has to be prepared in five days (*Pancharatrah*, five nights) duration. If *Dravadravya* was *Vrihi* and *Mamsa rasa* it has to be prepared within one day, two days for milk, three days for *Swarasa*, five days for *Takra, Aranala* etc., and twelve days for *Mula.*[9] Hence *Sneha Paka* was done in five days.

This heating was done in day time and allowed to cool in night time. Hence, author of Bhaishajya Ratnavali, Govindadas mentioned '*Ratrau'* (nights) term to highlight the importance of intermittent cooling.

During *Taila Paka*, vapours appeared due to boiling of *Aranala* and *Jambira.* During this phase, multiple readings of temperature, it showed around 90°c to 95°C. Water content (*Aranala* and *Jambira Swarasa*) reduced during the heating process due to

vaporization. Strong smell of *Kupilu* was observed at this stage with bitter/pungent aroma. It was unpleasant in nature. When maximum quantity of *Dravadravya* is reduced, *Kalka* start sticking to the bottom of the vessel. To avoid that continuous stirring was done. After this stage gradually the *Kalka* starts to float on the surface of the oil. *Snehasiddhi lakshanas* like appearance of *Kalka* in *Varti* form, *Phenodgama* etc was tested and got positive results in all the three samples. This shows that, preparation of oil was completed. After the oil preparation the aroma was acceptable. The colour of the *Taila* gradually changes during day by day. The end product of *Taila* was mustard brown in colour. The reason of this colour may be due to its processing in *Kupilu* and other ingredients. Different chemical changes occurs during the transferring of the properties from *Drava* and *Kalka* medium to *Taila* medium. For external application, *Taila* should be prepared either in *Madhyama* or *Kharapaka.* In this *Taila* preparation *mandagni*is maintained for all the three samples. This helps in easy evaporation of water and water-soluble extractives get slowly absorbed into the fat molecules.

Average Percentage of *Taila* obtained in three samples was around 90%. This is because of absorption of *Taila* in the *Kalkadravya.* The reason behind this was it is impossible to filter completely from *Kalka.* Some portion of *Taila* is consumed due to absorption of *Kalka.* Also, loss due to processing, filtration, adhering to the vessels etc. so for these reasons the lesser yield in *Taila* obtained. While preparing *Taila,* scholar experience the sensation of itching and dryness in throat region. Also increased feeling of thirst, mild itching sensation in skin region were observed. Hence specific precautions must be taken while preparing *Taila* like wearing gloves, mask because the fumes appearing while preparing *Taila* may cause irritation, dryness of mouth etc. Application of coconut oil/ moisturizer over skin region reduced the adverse effects. Likewise, direct contact also avoided.

Analytical Study

Analytical study provides the objective parameters to fix the standards for raw, processing and also finished products. To establish the basic standards, analytical study was necessary, so analytical study of *Vishatinduka Tailam* was done. According to CCRAS protocol for testing, the analytical study was carried out. Analytical parameters tested are refractive index, specific gravity, acid value, saponification value, iodine value, peroxide value and rancidity. *Taila* being a lipid module of dosage form has been investigated for rancidity by peroxide value, acid value and iodine value.

Organoleptic characteristics: The colour of the *Taila* was mustard yellow in initial stage turned into mustard brown due to content of *Vishatinduka* also due to processing over fire for more duration. The odour of the *Taila* was due to addition of *Jambeera* and *Aranala.* The aroma of the *Taila* is irritating over throat region and its increase the sensation of thirst. The consistency of *Vishatinduka Taila* is more viscous compared to *Tilataila* because of *Erandataila* presence.

Loss on drying is the loss expressed as percentage w/w resulting from water and volatile matter of any kind that can be driven off under specified conditions. Loss on drying 0% indicates that, no moisture content present in oil.

The acid value indicates the presence of free fatty acids in the oil sample. The free fatty acids are responsible for the rancidification of oil. Higher the fatty acids content of oils makes it rancid faster. Lesser percentage of fatty acid content makes the oil to rancid slower. Acid value 12.344mg/KOH/g at initial stage and in first month it was 12.90 and in 2nd month it was 12.90. Acid value was more in 1st and 2nd month implies that, more chances for rancidification. Due course of time, the rancidity of oil was increasing in nature. So, changes of unpleasant smell will get increased due to rancidity.

The iodine value indicates the degree of unsaturation of a fat or oil. It is defined as the number of grams of iodine absorbed by 100g of fat. Iodine value at initial stage and first month was 99.48 gl. In second month, it was 101.52. Iodine value was more in 2nd shelf-life period. The lesser the value shows the degree of unsaturation. The lesser unsaturation or in other words more saturation took place in 2nd shelf-life period when compared to 1st shelf life which signifies the more extractable constituents present in it. It also signifies the susceptible nature of shelf life to undergo rancid than the 1st shelf life greater the unsaturation leads to faster rancid. Iodine value of *Vishatinduka Tailam* sample was more in 2nd month. It denotes that unsaturation of fat/oil was more in 2nd month. The lesser unsaturation or in other words more saturation took place in 2nd month compared to 1st month, which signifies the more extractable constituents present in it.

The peroxide value indicates the degree of rancidification of oils. The increase value shows that the oil is turned rancid or spoiled. Also, it indicates the more chances of oxidation. Peroxide value is 5 mEq/kg, it was similar in all the month. As the normal peroxide value ranges in any oil is below 10 that is within the permissible limits. Neither of any of the sample crossed the limit. So, neither of samples got rancid.

Specific Gravity is the ratio of object density to that of water. It indicates the presence of solute content in the solvent. Here solvent is oil and solute refers to the extraction of active principles from the oil. Specific gravity of *Vishatinduka Tailam* was 0.932 in

initial stage, during shelf-life 1st month it was 0.932 and during 2nd month 0.930. The specific gravity of water is 1. The value greater than 1 will sink and lesser than 1 will float. Thus, in this study *Vishatinduka Tailam* was showing specific gravity lesser than 1. Therefore, its shows the ideal specific gravity of an oil.

Refractive index is the ratio of velocity of light in a vacuum to its velocity in a specified medium. It helps to identify the purity and measure the concentration of the substance. *Vishatinduka Tailam* during initial stage and 1st month was 73.5+_0.2% and 2nd month was 73.5+_0.1%. Approximately all the month refractive index was similar in nature.

Saponification value of oil or fat or ester is defined as the number of milligrams of KOH required to complete neutralize the free acids to saponify the ester present in 1gram of substances. It indicates the average molecular/chain length of all fatty acids present. *Vishatinduka Tailam* value= 112.22, 1st month shelf life= 117.83 and 2nd month shelf life=115.02. Saponification Value is inversely related to the chain length. Short chain has more absorption capacity. Higher the molecular weight of fat, smaller the saponification value. Low molecular weight fatty acids get quickly and easily absorbed. Here saponification value of 1st month shelf life is more compared to 2nd month shelf life, this indicates the presences of higher content of low molecular weight fatty acids. So *Vishatinduka Tailam* can be easily absorbed.

Rancidification is the process of complete or incomplete oxidation or hydrolysis of fats and oil when exposed to air, light or moisture or by bacteria, resulting in unpleasant taste and odour. Absence of rancidity in all month shows no spoilage of oil.

Density indicates weight of a given substance per volume of that substance. The density in initial and 1st month was 0.932g/cm^3. In 2nd month it was 0.930g/cm^3. The density is more in 1st month when compared to 2nd month. The density of *Taila* started to increase over the period of time. pH of 4.5+/_0.3 indicates *Vishatinduka Tailam* is acidic in nature.

Total fatty mater is defined as total amount of fatty mater, mostly fatty acids, that can be separated from a sample after splitting with mineral acid, usually HCl.1st month shelf life was 95.4% and 2nd month shelf life was 94.4%. It indicates that, more fat content present in 1st month.

Microbial contamination refers to the non-intended or accidental introduction of infectious material like bacteria, yeast, mould, fungi, virus, prions, protozoa or toxins and by products. Microbial contamination was nil in *Taila.* Absent of fungal content in oils showing the no fungal contamination of oil.

HPTLC: The HTPLC of *Vishatinduka Tailam* specific in the refractive domain done in n-butanol: acetic acid: water– 4:1:5 solvent system. HPTLC finger printing showed that the presence of 4 phytoconstituents. The standard Rf value of strychnine was 0.54 and brucine was 0.34.[10] In our study the Rf values are 0.37, 0.24, 0.11 and 0.04. The Rf value 0.37 was approximately equal to the value 0.34 of standard brucine. The remaining peaks may be complexes of strychnine and brucine. These metabolites were formed due to processing of oil and also due to interaction of various substances like, *Jambira, Aranala* etc. The phyto-constituents showing the qualitative effectiveness of *Vishatinduka Tailam.* By analysing the above parameters, it is evident that *Vishatinduka Tailam* was more stable and has good absorption power.

CONCLUSION

From the present study, it can be concluded from the analytical parameters, that, *Vishatinduka tailam* has stable nature with low molecular weight fatty acids. So *Vishatinduka Tailam* can be easily absorbed. This *Taila* was absence in fungal contamination. Saponification value shows the presence of long, medium and short chain fatty acids in *Vishatinduka Tailam* respectively suggesting its therapeutic utility in different routes of administration. Since *Vishatinduka Tailam* ingredients are effective in pain relief it can be applied externally in reduction of pain.

Figure 1: Ingredients of *Vishatinduka tailam*

Shodhitavishatindukabeeja

Vishatindukabeejachoorna

Aranala

Jambira

Tilataila

Eranda taila

Figure 2: Preparation of *Vishatinduka tailam*

Tilataila and Erandataila was poured

Vishatinduka kalka was added

Aranala was added

Jambiraswarasa was added

All ingredients are added and heated

Filtered taila

Figure 3: HPTLC Visualisations of sample

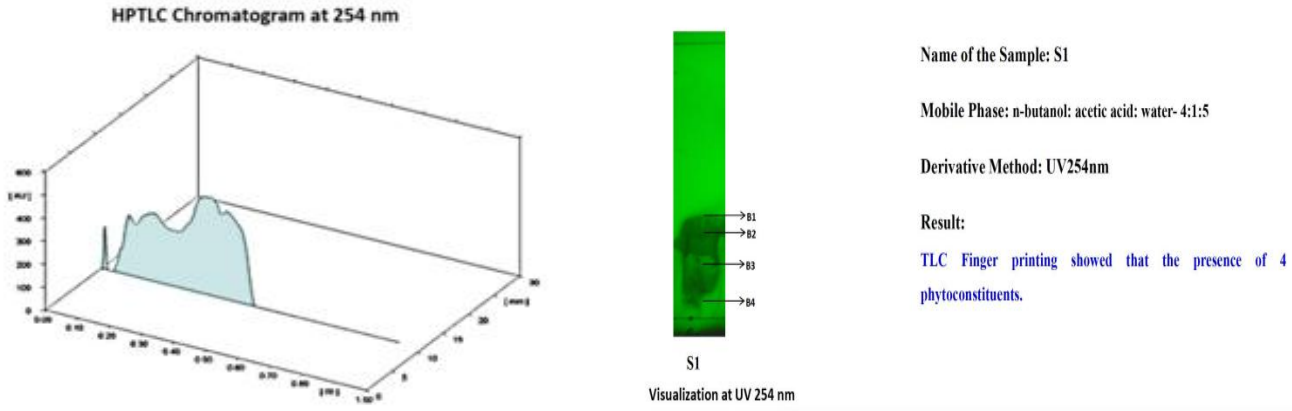
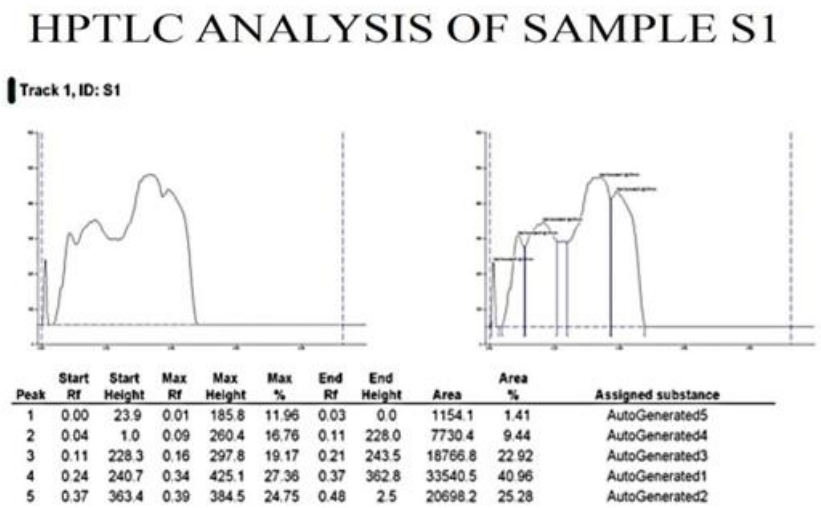


Figure 4: HPTLC Analysis of sample



Peak	Start Rf	Start Height	Max Rf	Max Height	Max %	End Rf	End Height	Area	Area %	Assigned substance
1	0.00	23.9	0.01	185.8	11.96	0.03	0.0	1154.1	1.41	AutoGenerated5
2	0.04	1.0	0.09	260.4	16.76	0.11	228.0	7730.4	9.44	AutoGenerated4
3	0.11	228.3	0.16	297.8	19.17	0.21	243.5	18766.8	22.92	AutoGenerated3
4	0.24	240.7	0.34	425.1	27.36	0.37	362.8	33540.5	40.96	AutoGenerated1
5	0.37	363.4	0.39	384.5	24.75	0.48	2.5	20698.2	25.28	AutoGenerated2

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***Address for correspondence**
Dr. Vanitha V
PG Scholar,
Department of Rasashastra and Bhaishajya Kalpana,
Sri Jayendra Saraswathi Ayurveda College and Hospital, Chennai-Bangalore Highway, Nazarathpet, Chennai, Tamilnadu, India.
Email:
Vanithavasanthan22@gmail.com