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# **RESEARCH ARTICLE**

## ANALYSIS OF BIOACTIVE COMPOUNDS FROM SINGLE HERBAL SIDDHA DISTILLATE "VASAMBU DRAVAGAM" THROUGH GC-MS

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<b>ARTICLE INFO</b>	ABSTRACT
Article History: Received 15 <sup>th</sup> February, 2018 Received in revised form 19 <sup>th</sup> March, 2018 Accepted 10 <sup>th</sup> April, 2018 Published online 30 <sup>th</sup> May, 2018	<b>Background:</b> <i>Theeneer</i> , the hydro distillate formulations of <i>Siddha</i> medicine is getting popularity and wide clinical acceptance owing to its faster therapeutic outcome. The raw drugs single or compound is preprocessed and subjected to distillation by following the traditional parameters mentioned in the siddha texts. <i>Vasambu</i> (Acoruscalamus Linn.) is a well known herb that has very reputed use in Traditional system like <i>Siddha</i> medicine and other Ethno medicinal practices. The rhizome part is distilled for its medicinal purposes and is termed as <i>Vasambu Dravagam</i> . The drug is indicated in <i>siddha</i> medicine for correcting the vititations of three humors namely. <i>Varhi</i> archel and <i>human</i> ( <i>Vatham</i> )
Key words:	pitham and Kapham).
Siddha medicine, Theeneer, Vasambu Dravagam, GC-MS, Asarone, Silicic acid.	<ul> <li>Objectives: The present study aims in preparing the distillate of Acoruscalamus rhizome and to analyze the drug with GC-MS (Gas chromatography Mass Spectrometry).</li> <li>Methods: The Rhizome part of <i>Vasambu</i> (Acoruscalamus Linn.) was distilled after following the preprocedures and further the distillate were analyzed for screening the active components.</li> <li>Results: <i>Vasambu Theeneer</i> was colorless with slight peculiar nauseating odor, and having mild sweet and pungent taste. Through GC-MS a total of 4 compounds were identified from Acorus distillate. Predominant peak belonging to 4-[4-(1-Phenyl-1H-1, 2, 4-triazol-5-yl)-1, 3-thiazol-2-yl] pyridine then asarone, Silicic acid and 2-(2-Benzothiazolyl)-3-[4-(dimethylamino) phenyl] acrylonitrile.</li> <li>Discussion: Asarone and the volatile siloxane silicic acid is the active component of this Acorus distillate which has reputed Pharmacological activity in deciding the outcome of the drug</li> <li>Conclusion: <i>Vasambu Dravagam</i> and its possibility in clinical aspects for broad spectrum of diseases may be supported with this preliminary screening and interpretations.</li> </ul>

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# INTRODUCTION

Hydro distillate formulations in *siddha* medicine are superior in terms of quality and potency. The terminology *Theeneer*<sup>(1)</sup>is attributed to extracts of herbal raw materials in aqueous nature with sharp potency. They are potential healers for specific ailments and some are used in a broad spectrum way to manage varieties of medical conditions. One of the fundamental principle by which the *siddha* system is established is the concept of *Mukutram*, the Trihumoral theory. A perfect physiological ratio of all the 3 humors namely *vazhi*, *azhal* and *iyyam* promote healthy well being of an individual free from diseases or degeneration<sup>(2)</sup>.

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Hundreds of diseases are categorized on the basis of deranged kutram (humors) or either due to its combined vitiations. It is the goal of the siddha practitioner to provide the best symptomatic relief, and simultaneously to treat the root cause of the disease, thereby to maintain the equilibrium of the trihumors. A drug which is capable of balancing all the three (MukutraSama Porul) is considered very superior and is prescribed as the finest supportive therapy along with other medications.<sup>(3)</sup> So many herbal distillates either single or compound formulations are been used in *siddha* practice with its notable indication or property in balancing the deranged humors. The distillate from the rhizome of Acoruscalamus is one such simple classical preparation. Vasambu (Acoruscalamus Linn.) belonging to family Araceae is a well known herb that has very reputed use in Traditional system like Siddhamedicine andother Ethnomedicinal practices.

Table. 1 General Description of Vasambu (4-8)

Color	Externally purplish brown to light brown Internally white		
Odor	Characteristic		
Suvai(Taste)	Karppu (Pungency)		
Thanmai (Potency)	Veppam (Hot)		
Pirivu (Division)	Karppu (Pungent)		
Chemical	Acorenone, isocalamendiol, Asarone, betasarone, monoterpene hydrocarbons, Sequestrine ketones, eugenol, isoeugenolmethylester,		
Constituents	Linalool, elemicin, acetophenone, dehydroabietic acid, a- Ursolic acid, Nonanoic acid, Fatty acids like oleic, linoleic, Linolenic acid		
	(George et al, 1986) palmitic, palmitoleic, stearic and myristic acids.		
Pharmacological	Anti-oxidant, Antiperiodic, Antifungal, Anthelmenthic, Carminative, Disinfectant, Emetic, Germicide, Insecticide, Sedative		
Actions	Stomachic, Stimulant, Tonics, Tranquilizer.		
Indications in	Toxic bites, pun (ulcers), Valippu (Convulsions), Gunmam (Gastro intestinal ailments), Rakthapitham (Bleeding disorders), Soolai		
Siddha Medicine	(Painful affections), Muppini (Delirium), Irumal (Cough), Pleehanoi (Diseases of Spleen), Silipatham (Elephantiasis), Kriminoi		
	(Helmenthiasis) Naakkupirazhchi (Stammering), Vayu (Gaseous disturbances) Seriyamai (Dyspepsia), Kazhichil (Diarrhoel diseases),		
	Sori Sirangu Kushttam (Scapies & other skin diseases) Muraikavchal (Periodic fever) etc		



Fig A. Raw Acoruscalamus



Fig. B. Period of soaking Fig. C. Distillation procedures



This perennial plant has creeping and extensively branched aromatic rhizome. Therapeutic potential is mainly attributed to its rhizome part. The rhizome part is distilled for its medicinal purposes and is termed as *Vasambu Dravagam* <sup>(9)</sup>. The drug is indicated for correcting the vitiations of three humors namely *Vazhi, azhal and Iyyam (Vatham, pitham and Kapham)* moreover the distillate is been traditionally used in *Siddha* practice for improving digestion, bowel regulation and for relieving flatus.

**Aim and objectives:** The present study aims in preparing the distillate of Acoruscalamus rhizome and to analyze the drug with GC-MS (Gas chromatography Mass Spectrometry).

The study may help to assess the efficacy of the drug for further clinical validations.

### **MATERIAL AND METHODS**

- A. Ingredient Details: Vasambu (dried rhizome of Acoruscalamus) (Fig. A)
- B. Method of Preparation of Distillate Sample (Fig. B & C) <sup>(9)</sup>: The dried rhizomes (300 g) were crushed well, mixed with 10 parts of pure water (3 liters) taken in a porcelain jar and soaked for a period of 3 days. On the 4<sup>th</sup> day the entire contents were transferred to a traditional still to initiate distillation. The distillate collected was preserved for analysis.

Peak	Retention	%Peak	Peak Intensity	Mol.	Name of the Compound	Chemical
no	Time	Area	Rank	Wt	-	Formula
1	8.25	13.71	2	208	Asarone	$C_{12}H_{16}O_3$
2	11.26	0.54	4	305	2-(2-Benzothiazolyl)-3-[4-(dimethylamino) phenyl] acrylonitrile.	$C_{18}H_{15}N_{3}S$
3	13.59	1.69	3	296	Silicic acid	C10H28O4Si3
4	15.16	84.06	1	305	4-[4-(1-Phenyl-1H-1,2,4-triazol-5-yl)-1,3-thiazol-2-yl]pyridine	$C_{16}H_{11}N_5S$

#### Table 2.Gas Chromatograph Report of Vasambu Dravagam.

### Table3. Bioactive compounds of Vasambu Dravagam





Fig. D. Gas Chromatography of Vasambu Dravagam

C. Gas Chromatography- MassSpectrometry (GC-MS) <sup>(10)</sup>:GC-MS is a sophisticated tool used in the separation, identification and quantitation of volatile and organic compounds present in the hydro distillate. GC-MS analysis of the distillate was performed with Agilent 7890B GC connected to 5977A MSD along with NIST Ver.2.1 MS data library Specification.

## RESULTS

*Vasambu Dravagam* was colorless with slight nauseating odor and mild sweet and pungent taste.

GC- MS reports of Vasambu Dravagam (Fig D, Table.2):The Gas chromatogram of Vasambu Dravagam revealed 4

prominent peaks with retention time ranging from 8.25 to 15.16. The detailed report of the compounds retention time (RT), Peak intensity rank, Molecular weight, name, chemical formula chemical structure and its pharmacological activities were presented in Table 2 & 3.

Table 3. Pharmacological significance of compounds spotted in  $Vasambu\ Dravagam^{(11-14)}$ 

Compounds		Activity
Asarone	1.	Anticonvulsant
	2.	Antipyretic
	3.	Anti Spasmodic
	4.	Cardio-depressant
	5.	CNS-Depressant
	6.	Cryoprotective.
	7.	Emetic
	8.	Fungicide
	9.	Myo relaxant
	10.	Hypothalamic Depressant
	11.	Insecticidal
	12.	Neuroprotective
	13.	Sedative
	14.	Tranquilizer
Silicic acid	1.	Anti diabetic

### DISCUSSION

An in depth knowledge of phytocomponents present in herbs is very much crucial for establishing the clinical significance of traditional formulations. The Analytical studies of Vasambu Dravagam the distillate prepared from the rhizome of Acoruscalamus shown 4 compounds and out of this Asarone and Silicic acid has wide report on its pharmacological activities. Asarone (2, 4, 5- trimethoxy -1 – propenyl benzene) is one of the principle bioactive constituent present in Acorus species especially in its rhizome. Asarone is also present as higher concentration in, Daucuscarota seeds, fruits of Piper cubeba and roots of sassafras albidum.<sup>(11)</sup> The intermediate compounds of asarone alpha and beta asarone have validated pharmacological activities. In a study the protective effect of alpha asarone were assessed against doxorubicin (DOX) induced nephritic syndrome in rats. The study revealed improvement in renal functions with simultaneous reduction in proteinuria, dyslipidemia and hypoalbuminemia. In addition to that an improved level of antioxidant activities was observed in renal tissues. The study supports its Nephroprotective, Hypolipidemic, Anti Inflammatory and Antioxidant activities (Sutariya and Saraf, 2018)<sup>(15)</sup>. In studies on standard animal seizure models alpha asaronol an active constituent derived from alpha Asarone demonstrated anticonvulsant and neuroprotective effects (Heet al., 2018). Various other Researches on alpha asarone indicate its anti nociceptive, Hypnotic and anxiolytic effects (Guiet al., 2018)<sup>(16)</sup>. While on the other intermediate the phenylpropanoid Beta asarone has been established for its Anti depressant, Anthelmenthic, antibacterial, anti anxiety, anti Alzheimer's, anti Parkinson's, anti epileptic, anti cancer, anti hyperlipidemic, antithrombotic, anticholestatic and radio protective properties (Chelianet al., 2017)<sup>(17)</sup>.Silicic acid is one of the forms of element silicon generally found in soils which is easily up taken by the plant roots (Heatheret al., 2007)<sup>(18)</sup>. Silicic acid is also named as orthosilicic acid is a common constituent present in so many herbal parts including leaves of Oryza sativa and Maclurapomifera<sup>(19)</sup>. These compounds exist as volatile siloxanes(Kathirvelet al., 2014)<sup>(20)</sup>in plant parts easily

extracted by hydro distillation. The drug may act in a multiple clinical associations or either expressed traditionally in terms of perfect balancer of health.

#### Conclusion

A herbal drug is a potential reservoir of thousands of active components. The extraction value of the constituents mainly depends on the solubility of the medium and the methods used during its preparation. Water soluble compounds are best extracted by soaking or boiling the raw drugs. Hydro distillates extract potent volatile and other organic compounds from the plant material. As a matter of fact It's inapplicable to isolate and study each component from a hydro distillate but the principle compounds which decides the outcome of its efficacy can be screened by sophisticated analytical techniques like GC-MS. We can predict the therapeutic potential of distillates to a greater extend by identifying such marker compounds and its intermediates or derivatives. From the study we may conclude the possibility of using the single herbal siddha distillate Vasambu Dravagam as a drug of choice for broadspectrum of clinical associated conditions. As GC-MS is only one of the analytical steps for the standardization of the drug, further studies have to be carried out for validating the safety and efficacy of this distillate.

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