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Pharmacognostic evaluation of a novel indigenous single herb formulation: *Peetha saireyaka* syrup

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ABSTRACT

Peetha saireyaka syrup is a single herb formulation, which was developed as a remedy for *Tundikeri* (Chronic Tonsillitis) based on traditional knowledge. The formulation is prepared with a herb root, *Peethasaireyaka (Barleria prionitis Linn.)*

Aim: To standardize *Peetha saireyaka* Syrup

Materials and methods: The raw materials were collected, authenticated & Physico - chemical studies like refractive index, specific gravity, viscosity, total solids, reducing and non-reducing sugar, and HPTLC were carried out as per the WHO guidelines, Indian Pharmacopoeia and Ayurvedic Pharmacopoeia

Conclusion: Standardization tests done on *Peetha saireyaka* syrup helped in authenticating and ensuring the quality of the same

Keywords: Syrup, Single herbal, Standardization, HPTLC, *Peetha saireyaka, Tundikeri*

INTRODUCTION

Ayurveda involves the use of natural elements to eliminate the root cause of the disease by restoring balance, at the same time create a healthy life-style to prevent the recurrence of imbalance. World Health Organization estimated that 80% of the word's inhabitants still rely mainly on traditional medicines for their health care. In *Ayurveda*, single or multiple herbs (poly herbal) are used for the treatment. Ayurveda has included herbs as one of its most powerful healing agents, which are recorded in the literature such as Vedas and Samhitas. Synthetic pharmaceuticals, are found out to be relatively more expensive and produce numerous undesirable side-effects despite their strong pharmacological action. Thus, people nowadays are shifting back to herbal medicines, which are originated from the nature and claim to be safer.[1]

Standardization of herbal medicines is the process of prescribing a set of standards or inherent characteristics, constant parameters, and definitive qualitative and quantitative values that carry an assurance of quality, efficacy, safety, and reproducibility[2]. The standardization of herbal medicines is always challenging as its medicines contain more than one active principle and the active compound is frequently unknown. The quality assessment of herbal formulations is important to justify their acceptability and safety.[3]

Peetha saireyaka syrup contains only a single herb root which is of *Peetha* (Yellow) variety of *Saireyaka*, used as folklore practice in coastal Karnataka and proved to be effective in the management of *Tundikeri* (Tonsillitis)[4]. The drug has *Tikta*

(Bitter)-Madhura (Sweet) Kinchit Amla Rasa (little sour), Usna Virya (hot in potency), Laghu Guna (Light) and Katu Vipaka (Pungent to digest), Jwarahara (reducing fever) and *Vranashodhana* (wound cleansing) properties, which helps in bringing down the cardinal symptoms of *Tundikeri* (Chronic tonsillitis) like *Shotha (swelling), Vidaha (Burning sensation), Kandu* (itching)etc[5]. The following table details the properties of the *Peetha saireyaka.*

Table 1: showing properties of ingredients of *Peetha saireyaka* syrup

Drug & Part Used	*Rasa*	*Guna*	*Veerya*	*Vipaka*	*Doshaghnatha*
Peetha saireyaka[6,7]. (Root)	*Tiktha, Madhura*	*Laghu*	*Ushna*	*Katu*	*Kapha vata hara*

MATERIALS AND METHODS

Physico - chemical studies like refractive index, specific gravity, viscosity, total solids, reducing and non-reducing sugar, and HPTLC were carried out as per the WHO guidelines, Indian Pharmacopoeia and Ayurvedic Pharmacopoeia.

Plant Material

Peetha saireyaka moola was collected from Davangere city of Karnataka state. The collected drug was identified and authenticated by Department of *Dravyaguna*, Sri Dharmasthala Manjunatheshwara College of Ayurveda & Hospital, Hassan – 573201, Karnataka.

Methodology

The studies were done at Sri Dharmasthala Manjunatheswara Centre for Research in Ayurveda and Allied Sciences, Kuthpady, Udupi, Karnataka State, India as per standard procedure.

Refractive index

Placed a drop of water on the prism and adjusted the drive knob in such a way that the boundary line intersects the separatrix exactly at the centre. Noted the reading. Distilled water has a refractive index of 1.3325 at 25˚C. The difference between the reading and 1.3325 gives the error of the instrument. If the reading is less than 1.3325, the error is minus (-) then the correction is plus (+) if the reading is more, the error is plus (+) and the correction is minus (-). Refractive index of oil is determined using 1 drop of the sample. The correction if any should be applied to the measured reading to get the accurate refractive index. Refractive index of the test samples was measured at 28˚C

Specific gravity

Cleaned a specific gravity bottle by shaking with acetone and then with ether. Dried the bottle and noted the weight. Cooled the sample solution to room Temperature. Carefully filled the specific gravity bottle with the test liquid, inserted the stopper and removed the surplus liquid. Noted the weight. Repeated the procedure using distilled water in place of sample solution.

Viscosity

The given sample is filled in a U tube viscometer in accordance with the expected viscosity of the liquid so that the fluid level stands within 0.2 mm of the filling mark of the viscometer when the capillary is vertical and the specified temperature is attained by the test liquid. The liquid is sucked or blown to the specified height of the viscometer and the time taken for the sample to pass the two marks is measured. Viscosity is measured using the formula

$$\eta 1 = \frac{\rho 1 t 1}{\rho 2 t 2} X \eta 2$$

η1 – Viscosity of sample

η2 - Viscosity of water

t1 and t 2- time taken for the sample and water to pass the meniscus

ρ1 and ρ2 – Density of sample and water

X= Specific gravity of sample x 0.9961/specific gravity of water

Π= Xx Time for samplex1.004/specific gravity of waterx70sec

Total solids

Transfer accurately weighed 50 g of the sample to an evaporating china dish, which have been dried to a constant weight and evaporate to dryness on a water bath, then dry at 105 degree Celsius for 3 hr. After cooling the dish containing the residue in a desiccator for 30 min, weigh it immediately. The weight of residue should comply with the requirements stated under the individual monograph.

Reducing and non-reducing sugar

10 g of sample was taken in a 250 ml volumetric flask and 200 ml of water was added. Slight excess solid basic Lead acetate was added to remove tannins and made up to the mark without disturbing the solution by adding water. Shake and filtered. Slight excess of solid Sodium oxalate was added to remove excess of basic Lead acetate, shake and filtered. This filtrate was used for the estimation of reducing sugar.

Reducing sugar: Take the sugar solution in a 50 ml burette.

Preliminary titration: 10 ml of Fehling's solution was pipette into a 250 ml conical flask, from the burette, 15 ml of the sugar solution was added. The liquid boiled on asbestos-covered gauze and further quantities of the sugar solution was added (One ml at a time) at 10 to 15 second intervals to the boiling liquid until the blue colour is nearly discharged. 3-5 drops of aqueous Methylene blue solution (1%) was added and continued the titration until the indicator is completely decolourised.

Accurate titration: The titration repeated, before heating, almost all of the sugar solution required to effect reduction of copper added. Gently boiled for two minutes. 3-5 drops of methylene blue indicator were added and the titration was completed within a total boiling time of three minutes. At the end point all the blue colour should be discharged and the liquid should be red.

The proportions of the various sugars, equivalent to 10 ml of Fehling's solution are taken from the table.

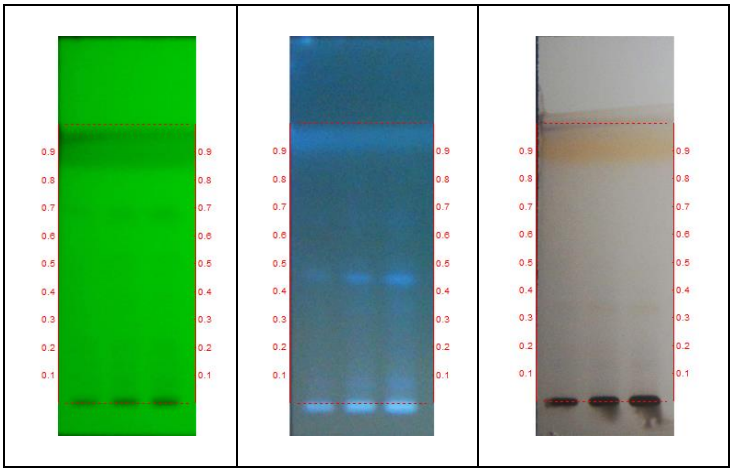
Total Sugar: 20ml of reducing sugar solution was taken and 10ml of Concentrated Hydrochloric acid added and kept it aside overnight.
Neutralized with approximately 1M Sodium hydroxide solution or with solid sodium carbonate and made up to 100 ml in a volumetric flask. Determined the total sugar content by the titrimetric method described above. Repeat the experiment twice and take the average value.

HPTLC

10ml of *Peetha saireyaka* syrup sample was partitioned with 20 ml butanol in a separating funnel and kept for 24hr. The butanol fraction was collected and filtered. The butanol was then made to evaporate on a water bath and it is dissolved in 10.0ml of methanol. 3, 6 and 9µl of the above sample were applied on a pre-coated silica gel F254 on aluminium plates to a band width of 7 mm using Lino mat 5 TLC applicator. The plate was developed in Toluene: Ethyl Acetate: Acetic acid: Methanol (3.0:3.0:0.8:0.2). The developed plates were visualized under short UV, long UV and after derivatisation with vanillin sulphuric acid spraying reagent, and scanned under UV 254nm, 366nm, and then derivatised plates were observed under white light. Rf, colour of the spots and densitometric scan were recorded.

Table 2: Results of standardization parameters

Parameter	***Results n = 3 % w/w***
	***Peetha saireyaka* syrup**
Specific gravity	1.1799
Refractive index	1.42532
Total solids	40.65
Total sugar	32.00
Reducing sugar	5.92
Non reducing sugar	26.08



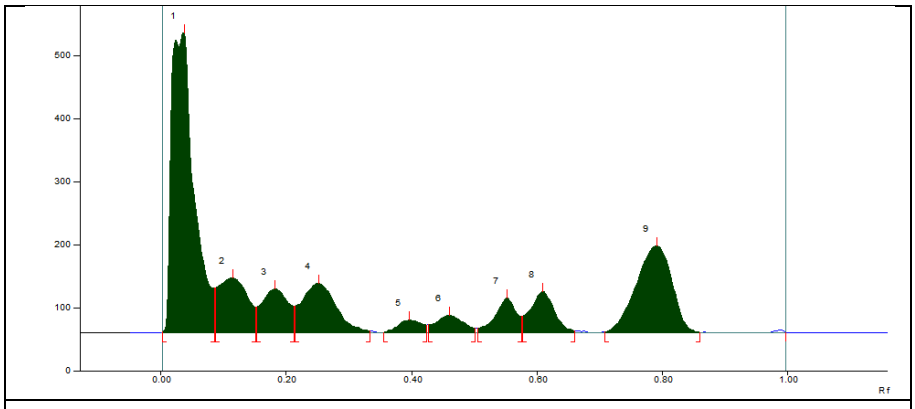
Track 1: *Peetha saireyaka syrup* - 3µl
Track 2: *Peetha saireyaka syrup* - 6µl
Track 3: *Peetha saireyaka syrup* - 9µl
Solvent system: Toluene: Ethyl aetate: Formic acid: Methanol (3:3: 0.8: 0.2)

Fig 1: HPTLC Photo documentation of sample of *Peetha saireyaka syrup*

Table 3: Rf values of sample of *Peetha saireyaka* *syrup*

Short UV	**Long UV**	**Post derivatisation**
0.08 (L. green)	0.08 (F. Blue)	-
-	-	0.14 (Purple)
0.20 (L. green)	-	0.19 (Purple)
-	0.34 (F. Blue)	0.34 (Yellow)
-	0.44 (F. Blue)	-
-	0.65 (F. Blue)	-
0.68 (L. green)	-	-

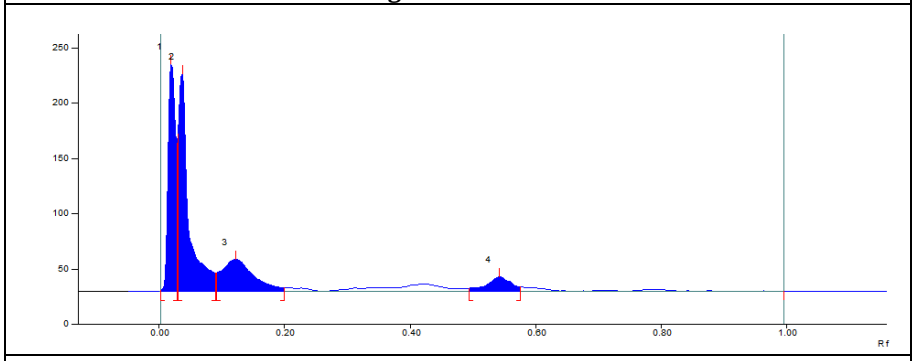
**D – dark; L – light; F – fluorescent*



Track 3, ID: Peethasaireyaka syrup

Peak	Start Position	Start Height	Max Position	Max Height	Max %	End Position	End Height	Area	Area %
1	0.00 Rf	1.6 AU	0.04 Rf	474.5 AU	46.73 %	0.09 Rf	70.7 AU	12252.3 AU	40.18 %
2	0.09 Rf	71.2 AU	0.11 Rf	86.8 AU	8.55 %	0.15 Rf	40.7 AU	2943.7 AU	9.65 %
3	0.15 Rf	40.9 AU	0.18 Rf	69.8 AU	6.88 %	0.21 Rf	41.9 AU	2111.1 AU	6.92 %
4	0.21 Rf	42.2 AU	0.25 Rf	78.1 AU	7.70 %	0.33 Rf	2.7 AU	3058.2 AU	10.03 %
5	0.36 Rf	0.3 AU	0.40 Rf	20.3 AU	2.00 %	0.42 Rf	12.3 AU	548.4 AU	1.80 %
6	0.43 Rf	12.4 AU	0.46 Rf	28.0 AU	2.76 %	0.50 Rf	7.2 AU	864.7 AU	2.84 %
7	0.51 Rf	7.4 AU	0.55 Rf	55.1 AU	5.42 %	0.58 Rf	25.9 AU	1298.1 AU	4.26 %
8	0.58 Rf	26.3 AU	0.61 Rf	65.8 AU	6.48 %	0.66 Rf	3.2 AU	1792.6 AU	5.88 %
9	0.71 Rf	1.1 AU	0.79 Rf	136.9 AU	13.49 %	0.86 Rf	0.7 AU	5625.2 AU	18.45 %

Figure at 254nm



Track 3, ID: Peethasaireyaka syrup

Peak	Start Position	Start Height	Max Position	Max Height	Max %	End Position	End Height	Area	Area %
1	0.00 Rf	0.6 AU	0.02 Rf	206.0 AU	46.26 %	0.03 Rf	33.6 AU	1758.6 AU	32.13 %
2	0.03 Rf	142.3 AU	0.04 Rf	197.0 AU	44.25 %	0.09 Rf	16.9 AU	2295.2 AU	41.93 %
3	0.09 Rf	16.9 AU	0.12 Rf	29.0 AU	6.52 %	0.20 Rf	3.4 AU	1057.7 AU	19.32 %
4	0.50 Rf	2.9 AU	0.54 Rf	13.2 AU	2.96 %	0.58 Rf	4.2 AU	362.1 AU	6.62 %

Figure at 366nm

Fig 2: Densitometric scan of the sample of *Peetha saireyaka* *syrup*

RESULTS & DISCUSSION

The standardization parameters of *Peetha saireyaka* syrup are detailed in Table 2. The TLC photo documentation of *Peetha saireyaka syrup* is shown in Fig. 1. The Rf values of sample of *Peetha saireyaka syrup* is detailed in Table 3. The Densitometric Scan of *Peetha saireyaka* syrup is shown in Fig. 2. The physicochemical standards would serve as preliminary test for the standardization of the formulation. Tests such as refractive index, specific gravity, viscosity, total solids, reducing and non-reducing sugar, and HPTLC, results of TLC photo documentation, the unique Rf values, densitometric scan and densitogram obtained at different wavelengths can be used as fingerprint to identify the herbal drug formulation, *Peetha saireyaka* syrup.

CONCLUSION

Peetha saireyaka syrup has been standardized using diverse scientific quality parameters. The results obtained can be used as reference while setting the pharmacopoeial standards for *Peetha saireyaka syrup* for the benefit of the end user without any unwarranted complications.

CONFLICT OF INTEREST

Nil

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