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Standardisation Study of Kwatha Cumas

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Abstract The present paper deals with the standardization of kwatha cumas such as dhanyapanchak kwatha cuma, guduchyadigana kwatha cuma and stanyajanankashaya cuma. These are the important Ayurvedic formulations used for peri-natal care of mother and child health. Standardization of kwatha cumas were achieved by physico-chemical analysis, qualitative inorganic and organic analysis, thin layer chromatography (TLC), UV- visible spectrophotometry and high performance liquid chromatographic (HPLC) fingerprint studies. TLC study of kwatha cumas was carried out in Ethyl acetate: Methanol: Water solvent system. Ethanol extracts of kwatha cumas were used for UV- visible spectrophotometry and qualitative HPLC fingerprint study.

Key words Standardization, Kwatha cumas, TLC and HPLC.

Introduction

Kwatha cumas are compound coarse powders, which are intended for usage whenever particular decoction is required. In the present paper, three-kwatha cumas viz., dhanyapachak kwatha cuma, guduchyadigana kwatha cuma and stanyajanana kashaya cuma were selected for standardization study. These kwatha cumas are used for the perinatal care of mother and child health. Process standardization of kwatha cumas namely, arkadi kwatha, phalatrikadi kwatha and rasna saptaka kwatha cumas has been reported. According to the analytical data, TLC and organic qualitative analysis, the preparation by the second method (by individual powdering of ingredients and compounding them in required proportion to get the final product) revealed more active principles¹.

The clinical study of dhanyapanchak kashaya cuma showed that this herbal drug has beneficial effect on dyspeptic symptoms².

Materials and Methods

The authentic ingredients were procured from the local market of Hyderabad, Andhra Pradesh and were botanically identified. Dhanyapanchak kwatha cuma, guduchyadigana kwatha cuma and stanyajanana kashaya cuma were prepared as per the procedure described in Ayurvedic Formulary of India^{3&4}.

Analytical study

The prepared samples were analyzed for the parameters such as pH (1% aqueous), moisture content, total ash, acid insoluble ash, alcohol soluble extractive, water-soluble extractive, qualitative inorganic and organic analysis^{5&6}.

Thin layer chromatography

TLC plates were prepared as per the procedure described by Stahl⁷. The 4% alcoholic extracts of the samples were prepared by soaking them for 18h in alcohol. Alcoholic extracts were filtered and about 100µl was loaded on the TLC plate and eluted in ethyl acetate: methanol: water (100:13.5:10) solvent system⁸. The plates were sprayed with vanillin-sulphuric acid reagent and the spots were detected after heating at 110°C for 30min. Rf value of each spot was calculated.

Sample preparation

The 4% alcoholic extracts of the samples were prepared by soaking them for 18h in alcohol. The extracts were filtered through Whatman filter paper number 1 using high-pressure vacuum pump. The samples were used for UV- visible spectrophotometric and HPLC fingerprint study.

UV-visible spectrophotometric analysis

The samples were scanned over a range of 200-800nm using ELICO (SL-159) UV-visible spectrophotometer equipped with quartz cuvettes of 10mm path length and UV-visible spectrasoft software. Alcohol was used as a reference.

HPLC analysis

A gradient HPLC (Shimadzu HPLC Class VP series) with two LC- 10 AT VP pumps (Shimadzu), variable wavelength programmable photo diode array detector SPD-M10A VP (Shimadzu), CTO-10AS VP column oven (Shimadzu), SCL-10A VP system controller (Shimadzu) and reverse phase Luna 5µ C₁₈ (2) Phenomenex column (250mm X 4.6mm) was used. The HPLC system was equipped with software Class VP series version 6.1 (Shimadzu). The mobile phase components methanol: water were filtered through 0.2µ membrane filter before use and pumped from the solvent reservoir to the column at a flow rate of 1ml/min which yielded a column back pressure of 220 kgf/cm². The column temperature was maintained at 27°C. 20µl of sample was injected by using Rheodyne syringe (Model 7202, Hamilton).

Results and Discussion

The data of physico-chemical analysis of dhanyapanchak kwatha curna, guduchyadigana kwatha curna and stanyajanana kashaya curna is summarized in Table-1. The data of qualitative inorganic and organic analysis of kwatha curnas is summarized in Table-2. The data of TLC study of kwatha curnas is summarized in Table-3. The UV-visible spectrophotometric data of kwatha curnas is tabulated in Table-4.

Table 1. Analytical data of kwatha curnas

S. No.	Analytical Parameters	Dhanyapanchak kwatha curna	Guduchyadigana kwatha curna	Stanyajanana kwatha curna
1.	pH (1% aqueous)	5.02	5.77	6.26
2.	Moisture (%)	8.08	6.65	6.7
3.	Total ash (%)	7.11	7.92	18
4.	Acid insoluble ash (%)	2.03	0.17	10.65
5.	Alcohol soluble extractive (%)	7.65	6.15	3.5
6.	Water soluble extractive (%)	13.55	6.1	4.5

Table 2. Qualitative inorganic and organic analytical data of kwatha cumas

S. No.	Analytical Parameters	Dhanyapanchak kwatha cuma	Guducyadigana kwatha cuma	Stanyajanana kwatha cuma
1.	Calcium	+ve	+ve	+ve
2.	Magnesium	+ve	+ve	+ve
3.	Iron	+ve	+ve	+ve
4.	Aluminum	-ve	-ve	+ve
5.	Lead	-ve	-ve	-ve
6.	Arsenic	-ve	-ve	-ve
7.	Mercury	-ve	-ve	-ve
8.	Alkaloids	+ve	+ve	-ve
9.	Steroids	+ve	+ve	-ve
10.	Phenols	+ve	+ve	+ve
11.	Tannins	+ve	+ve	+ve
12.	Glycosides	+ve	+ve	+ve
13.	Resins	+ve	+ve	+ve
14.	Saponins	+ve	+ve	+ve
15.	Flavonoids	+ve	+ve	-ve

Table 3. Data of TLC study of kwatha cumas

S. No.	Dhanyapanchak kwatha cuma Rf value (color)	Guducyadigana kwatha cuma Rf value (color)	Stanyajanana kwatha cuma Rf value (color)
1.	0.07(BG)	0.08(BG)	0.07(BG)
2.	0.17(LBG)	0.19(LBG)	0.22(LBG)
3.	0.24(BG)	0.25(LBG)	0.30(LBG)
4.	0.97(V)	0.97(V)	0.97(V)

Where, V- Violet, BG – Blue Green, LBG – Light Blue Green

Table 4. Data of UV-Visible Spectrophotometric study of Kwatha Cumas

S. No.	Dhanyapanchak kwatha cuma		Guducyadigana kwatha cuma		Stanyajanana kwatha cuma	
	W.L (nm)	O.D.	W.L (nm)	O.D.	W.L (nm)	O.D.
1	265	2.799	270	2.686	297	2.670
2	300	2.829	299	2.716		
3	464	3.088	390	2.102		
4	489	3.268				
5	664	0.283				

Where, W.L- Wavelength (nm) O.D.- Optical density

The qualitative HPLC fingerprint profiles of dhanyapanchak kwatha cuma, guducyadigana kwatha cuma and stanyajanana kashaya cuma are shown in Figure – 1 to 3 respectively. The HPLC chromatogram of dhanyapanchak kwatha cuma showed three prominent peaks at a retention time of

2.763min, 3.328min and 24.245min with an area percentage of 27.55, 6.17 and 24.36. The HPLC chromatogram of guduchyadigana kwatha cuma showed three prominent peaks at a retention time of 2.848min, 3.328min and 22.603min with an area percentage of 46.77, 13.24 and 13.86. The HPLC chromatogram of stanyajanana kashaya cuma showed ten prominent peaks at a retention time of 2.880min, 3.339min, 6.816min, 13.621min, 15.477min, 17.291min, 18.069min, 25.013min, 28.555min and 29.323min with an area percentage of 2.57, 9.03, 3.72, 2.99, 3.19, 2.48, 3.09, 11.74, 1.97 and 1.27.

Figure 1. HPLC Chromatogram of Dhanyapanchak Kwatha Cuma

Figure 2. HPLC Chromatogram of Guduchyandigana Kwatha Cuma

Figure 3. HPLC Chromatogram of Stanyajanana Kashaya Cuma

Conclusion

The analytical data, TLC, UV-visible spectrophotometric and HPLC fingerprint profiles evolved can be considered as viable parameters which will go a long way for prescribing a dependable standards to these preparations.

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