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Research Article

PHARMACOGNOSTICAL CHARACTERIZATION OF AN AYURVEDIC POWDERED FORMULATION: PANCHSAKAR CHURNA

P. Kumar[1]*, S. Jha[2] and T.Naved[3]

[1]Ram-Eesh Institute of Vocational and Technical Education, Greater Noida, Uttar Pradesh, India.
[2]Department of Pharmaceutical Sciences, Birla Institute of Technology, Mesra, Ranchi, Jharkhand, India.
[3]Amity Institute of Pharmacy, Amity University, Noida, Uttar Pradesh, India.

***Corresponding Author: puspendrapatel9@gmail.com**

ABSTRACT

Most of the traditional systems of medicines are effective but the need is just to validate them to assess the quality,quantity and purity of the drugs. In this direction a polyherbal Ayurvedic formulation Panchsakar churna used in constipation and piles was taken for pharmacognostical characterization. Panchsakar churna is the composition of *Cassia angustifolia* Vahl. leaf, *Terminalia chebula* Retz. fruit, *Zingiber officinale* Rosc. rhizome, *Foeniculum vulgare* Mill. fruit and Saindhava lavana. In-house preparation and three marketed samples have been pharmacognostically characterized by microscopy and physical constant values. Microscopy was performed by Leica microscope for identification of different diagnostic characters of the ingredients and formulatons. Anatomical study of each ingredient of the formulation and quantitative microscopy of *Cassia angustifolia* Vahl. leaf was performed for the authentication of the senna leaf. Study of Physical constant values was performed for all the formulation and ingredients and compared with the available standard values of the ingredients. Thus the above parameters were useful for the Pharmacognostical characterization of the Panchsakar churna and its ingredients. The set parameters were found to be simple to evaluate the churna and can be used as reference standards for the quality control / quality assurance of a Polyherbal powder formulation.

Keywords: Panchsakar churna, Standardization, Pharmacognostical, Ash values.

INTRODUCTION

According to an estimate of W.H.O nearly 80% populations of developing countries relies on traditional medicines, mostly on plant drugs for their primary health care needs[2]. Most of the traditional systems of medicines are effective but the need is just to validate them. Panchsakar churna is an Ayurvedic proprietary medicine mentioned in the Ayurveda Sarsangraha. This churna is used in constipation, piles and other abdominal diseases. Panchsakar churna is the composition of one part for each ingredient Cassia angustifolia Vahl. leaf, Terminalia

chebula Retz. fruit, Zingiber officinale Rosc. rhizome, Foeniculum vulgare Mill. fruit and Saindhava lavana.

For present study we have prepared in-house Panchsakar (IH) and selected three marketed Panchsakar (M1,M2 and M3), we have pharmacognostically characterized the individual constituents of Panchsakar churna with respect to anatomical studies, physical constant values (i.e. Extractive value, Total Ash value, Water soluble / Water insoluble ash value, Acid soluble Ash value, Moisture content) and quantitative microscopy (i.e. Stomatal index, Palisade ratio, Vein islet number and Vein termination number) of *Cassia angustifolia* Vahl. leaf was performed for the authentication of the senna leaf.

MATERIALS AND METHODS

Plant Materials

The plant materials of Panchsakar churna were collected from the different sources. Senna (*Cassia angustifolia* Vahl.) was purchased from K. Mohamad and co., Tinnevally, Tamilnadu (India); Haritaki (*Terminalia chebula* Retz.) was collected from the Raigarh district of Chhatisgarh(India); Ginger (*Zingiber officinale* Rosc.) was collected from the Bilaspur district of Chhatisgarh(India); Fennel (*Foeniculum vulgare* Mill.) and Saindhav lavana was purchased from the local market of Ranchi, Jharkhand (India). All ingredients were authenticated by Dr. S. Jha., Associate Professor, Department of Pharmaceutical sciences, B.I.T Mesra, Ranchi. Parts of the ingredients was crushed to powder using grinder and passed through sieve number# 85. In-house Panchsakar churna was prepared from these powders by mixing them in one part for each ingredient. Marketed Panchsakar churna supplied from three different companies were also procured and named as M1, M2 and M3.

Anatomical study of each ingredient of Panchsakar Churna

For Anatomical study invariably slides were prepared. A transverse section of required part of ingredient was taken on a glass slide to which are added a few drops of saffranin for 1-2 minute then washed with water. Then mounted with glycerin. Care however is to be taken to avoid air bubbles and to see that there is sufficient glycerine under the cover slip. Excess of glycerine outside the cover slip is to be withdrawn using a blotting paper. Then the anatomy of the transverse section were observed and identified under Leica microscope (10 x & 40x). [6] Anatomical study for each ingredients of Panchsakar Churna transverse section of midrib of *Cassia angustifolia* (Fig.1) *Foeniculum vulgare* fruit (Fig.2) Zingiber *officinale* rhizome (Fig.3) and *Terminalia chebula* fruit (Fig.4) were done.

Powder microscopy of Panchsakar Churna

A judicious quantity of powder was taken on a glass slide to which was added a few drops of chloral hydrate and was heated for 1-2 minute after placing a cover slip, care should be taken to avoid air bubbles and to see that there was sufficient chloral hydrate under the cover slip. Excess of chloral hydrate outside the cover slip is to be withdrawn using a blotting paper. (Chloral hydrate is used to clear the tissues and to bring in clarity of the view). Lignified tissues are to be confirmed by staining. To the powder a few drops of mixture of 1:1 Phloroglucinol +Conc.HCl were added after 3 to 4 minutes, it was finally mounted in chloral hydrate / glycerine and observed under Leica microscope (10 x & 40x) Powder microscopies of each ingredients of Panchsakar Churna were done and unique identifying characters were studied i.e. Unicellular trichome of *C.angustifolia* (Fig.5) Endosperm of *Foeniculum vulgare* (Fig.6) Stach grains of *Zingiber officinale* (Fig.7) and Stone cell of *Terminalia chebula* (Fig. 8).

Quantitative microscopy of *Cassia angustifolia* leaf

Determination of Stomatal index of *Cassia angustifolia* Vahl. Leaf

The leaf was teared in such a way that both the upper and lower epidermis was separated. A drop of chloral hydrate was added to the peeled or teared leaf part and then heated for few seconds. A drop of methanol was added after five minutes washed with water. A drop of safranin was added and washed immediately with water twice or thrice. The stained portion was kept in a slide and then mounted with glycerin. Then by using stage micrometer and camera lucida, one square mm was drawn in a black sheet. Then by the

help of microscope (10x40*x*) and camera lucida the number of stomata and epidermal cells were counted within the square. Those cells were not counted more than half portions of which were outside the square. Ten observations were done to measure Stomatal index of both upper and lower epidermis. [10]

Determination of Palisade ratio of Cassia *angustifolia* Vahl. Leaf

Small pieces of leaves from the base, middle and apex position of lamina were taken. Leaf pieces were boiled in conc. Chloral hydrate solution by placing in test tubes. The test tubes were kept in water bath till the leaf pieces become chlorophyll free. Then by using camera lucida, four adjacent cells of upper epidermis were traced. Then focused on the palisade layer and traced off the palisade cells beneath the four epidermal cells which were already traced. The palisade cells which were outside more than half portion were not counted. 24 observations were. [10]

Determination of Vein islet number and vein termination number of *Cassia angustifolia* Vahl. Leaf

A few cut portions of the leaf from the central region of lamina of 4 mm^2 were boiled in conc. Chloral hydrate solution by placing in test tubes. The test tubes were kept in water bath till the leaf pieces become transparent. Then each portion was kept on a slide with lower portion placing upward so that vein were prominent on the lower surface and a small drop of glycerin was added. 5*x* eyepiece and low power objective 10*x* were used. Stage micrometer was focused and camera lucida was fixed. A drawing sheet was placed on the side of the microscope where camera lucida was fixed. Then using stage micrometer 1 mm sq. was drawn. Image of the leaf was made to super impose the square on the drawing sheet. Vein islet were traced and counted. 10 observations were done to measure the vein islet number and vein termination number[8].

Determination of Extractive Values

Determination of Alcohol soluble Extractives

5g of air dried Powder of ingredients of Panchsakar churna and Panchsakar churna formulations were taken and macerated with 100ml of methanol of the specified strength (95%) in a closed flask for 24 hours, shaking frequently for the first 6 hrs and then was allowed to stand for 18 hrs. It was then filtered taking precautions against loss of methanol. 25ml of the filtrate was evaporated to dryness in a tarred flat bottomed shallow dish and was then dried at 105°C and weighed. [9] The percentage of the methanol soluble extractives was calculated with reference to air dried Powder of ingredients of Panchsakar churna and Panchsakar churna formulations. (Table 1)

Determination of Water soluble Extractives

5g of air dried Powder of ingredients of Panchsakar churna and Panchsakar churna formulations were taken and macerated with 100ml of chloroform water (0.25ml of chloroform with volume made up to 100ml with distilled water). In a closed flask for 24 hours, shaking frequently for the first 6 hrs and then was allowed to stand for 18 hrs. It was then filtered taking precautions against loss of chloroform water. 25ml of the filtrate was evaporated to dryness in a tarred flat bottomed shallow dish and was then dried at 105°C and weighed.[9] The percentage of the water soluble extractives was calculated with reference to air dried Powder of ingredients of Panchsakar churna and Panchsakar churna formulations. (Table 2).

Determination of Moisture Content

Moisture content determination was done by IR moisture balance. In this method sufficient amount of powder drug is spread in the evaporating plate of IR moisture balance till the pointer touches zero. Then temperature is maintained at 105^0 for 30 minute, after that moisture content was measured directly in percentage. (Table 3).

Determination of Ash values

Determination of total Ash values

3g of air dried Powder of ingredients of Panchsakar churna and Panchsakar churna formulations were taken in a tarred silica dish and were incinerated at a temperature not exceeding 450°C until free from carbon, and it was then cooled and weighed. [9] The percentage of ash was calculated with reference to air dried Powder of ingredients of Panchsakar churna and Panchsakar churna formulations. (Table 4).

Determination of water soluble and water insoluble ash values

The total ash obtained from above section was boiled with 25 ml water for five minutes and then filter through an ash less filter paper (whatmann#1). The filter paper was ignited in the silica crucible. Then cooled and the water insoluble ash was weighed. The water soluble ash was calculated by subtracting the water insoluble ash from the total ash. [9] Then the percentage of water soluble ash was calculated with reference to the air dried drug. (Table 3).

Determination Acid insoluble ash values

About 3g of the powdered drug was accurately weighed in a tarred silica crucible. Incinerate the powdered drug by gradually increasing the heat until free from carbon and cooled before weighing. The ignition was repeated until constant weight was observed. The total ash obtained was boiled for five minutes with 25 ml of 6N HCl and then filtered through ash less filter paper (whatmann#1).The filter paper was ignited in the silica crucible, cooled and acid soluble ash was calculated by weighing[9]. (Table 6)

RESULTS AND DISCUSSION

Anatomical studies of the each ingredient are showing same anatomical characteristics of the selected ingredients. Powder microscopy of the formulations shows that presence of each ingredient in the all formulations. Stomatal index of upper epidermis, Stomatal index of lower epidermis, Palisade ratio, Vein islet number and Vein termination number of *Cassia angustifolia* Vahl leaf were found to be 17.24-20.00; 16.98-19.29; 4.00-7.25; 19-22; 24-33 respectively, which are between the reference ranges. Extractive values of the ingredients were passed the Ayurvedic pharmacopoeial standard, while formulation M2 shows the maximum extractive values. Moisture content values of the ingredients passed the Indian pharmacopoeial standard. Ash values of the ingredients passed the Ayurvedic pharmacopoeial standard, while formulation M3 shows the least total ash value.

CONCLUSIONS

The study shows that all ingredients were genuine and passed the microscopical studied including quantitative microscopy. Ash values, Extractive values and moisture content values were passed the Ayurvedic pharmacopoeial standard for ingredient, while all these standards were established for all the formulations.

Table 1: Methanol soluble extractive values of various ingredients of Panchsakar churna and Formulations

S. No	Sample	Extractive Value (Methanol %w/w) ±SEM	Reference Value (Ayurvedic Pharmacopoeia)
1	*C. angustifolia*	17.6±1.313	≥ 3%
2	*T. chebula*	62.4±1.131	≥ 40%
3	*Z. officinale*	7.60±0.566	≥ 3%
4	*F. vulgare*	8.80±0.566	≥ 4%
5	*Formulation IH*	30.0±0.566	-
6	*Formulation M1*	31.2±1.313	-
7	*Formulation M2*	34.8±0.566	-
8	*Formulation M3*	27.2±0.000	-

Table 2: Water soluble extractive values of various ingredients of Panchsakar churna and Formulations

S. No.	*Sample*	Extractive Value (Methanol %w/w) ±SEM	Reference Value (Ayurvedic Pharmacopoeia)
1	*C. angustifolia*	30.0±0.566	≥ 25%
2	*T. chebula*	62.8±0.566	≥ 60%
3	*Z. officinale*	14.8±0.566	≥ 10%
4	*F. vulgare*	6.80±0.566	≥ 1%
5	*Formulation IH*	38.4±1.131	-
6	*Formulation M1*	35.6±1.697	-
7	*Formulation M2*	40.0±1.131	-
8	*Formulation M3*	34.8±1.697	-

Table 3: Moisture content of various ingredients of Panchsakar churna and Formulations

S. No.	Sample	Moisture content (%w/w)	Reference value (%w/w) (Indian Pharmacopoeia, 2007)
1	*C. angustifolia*	3.8	≤12
2	*T. chebula*	2.6	≤12
3	*Z. officinale*	4.0	≤12
4	*F. vulgare*	2.8	-
5	*Formulation IH*	2.6	-
6	*Formulation M1*	2.6	-
7	*Formulation M2*	3.2	-
8	*Formulation M3*	2.8	-

Table 4: Total Ash values of various ingredients of Panchsakar churna and Formulations

S.No.	Sample	Ash value (%w/w) ±SEM	Reference Value (Ayurvedic Pharmacopoeia)
1	*C. angustifolia*	9.295±0.120	≤14%
2	*T. chebula*	3.125±0.629	≤5%
3	*Z. officinale*	5.780±0.170	≤6%
4	*F. vulgare*	6.005±0.714	≤12%
5	*Formulation IH*	22.93±0.170	-
6	*Formulation M1*	23.77±0.534	-
7	*Formulation M2*	24.09±0.559	-
8	*Formulation M3*	20.85±0.304	-

Table 5: Water soluble/Water insoluble Ash values of various ingredients of Panchsakar churna and Formulations

S. No.	Sample	Water insoluble ash value (%w/w)	Water soluble ash value (%w/w)
1	*C. angustifolia*	7.410	1.970
2	*T. chebula*	0.793	2.776
3	*Z. officinale*	3.780	1.880
4	*F. vulgare*	3.090	2.413
5	*Formulation IH*	4.767	18.04*****
6	*Formulation M1*	5.003	19.14*****
7	*Formulation M2*	4.990	18.70*****
8	*Formulation M3*	4.343	16.72*****

***** Water soluble ash value was increased due to presence of Sandhav lavana in the Panchsakar Churna.

Table 6: Acid insoluble Ash values of various ingredients of Panchsakar churna and Formulations

S. No.	Sample	Acid insoluble ash value	Reference Value (Ayurvedic Pharmacopoeia)
1	*C. angustifolia*	1.080	≤2%
2	*T. chebula*	2.423	≤5%
3	*Z. officinale*	1.553	-
4	*F. vulgare*	1.477	≤1.5%
5	*Formulation IH*	1.450	-
6	*Formulation M1*	1.790	-
7	*Formulation M2*	1.710	-
8	*Formulation M3*	1.427	-

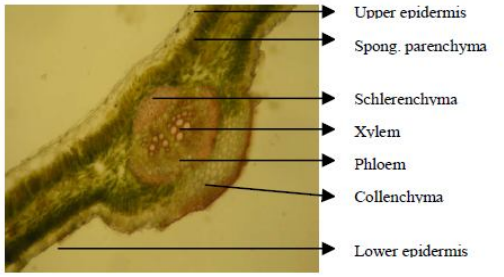

Figure 1: T.S. of midrib of ***Cassia angustifolia***

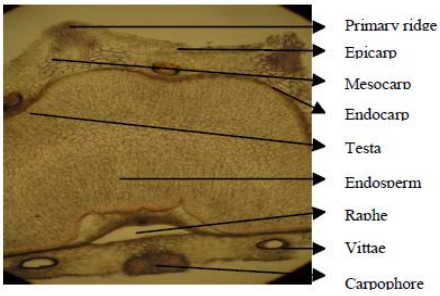

Figure 2: T.S. of ***Foeniculum vulgare*** fruit.

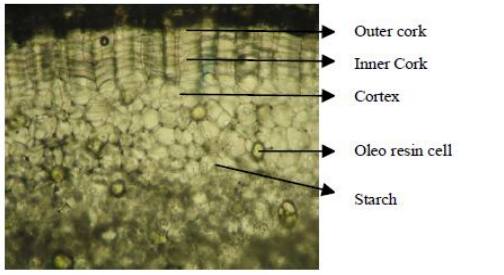

Figure 3: T.S. of Zingiber ***officinale*** rhizome

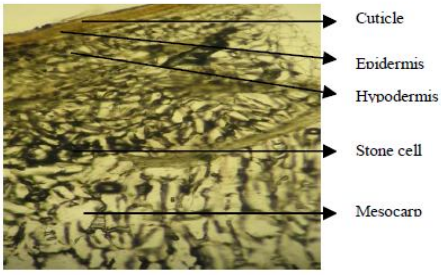

Figure 4: T.S. of ***Terminalia chebula*** fruit.

Figure 5:Unicellular trichome of *C.angustifolia*

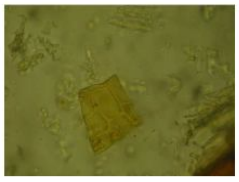

Figure 6: Endosperm of *Foeniculum vulgare*

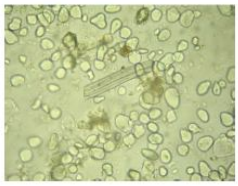

Figure 7: Stach grains of *Zingiber officinale.*

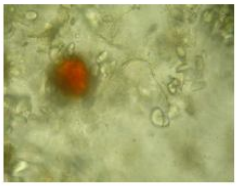

Figure 8: Stone cell of *Terminalia chebula.*

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