RESEARCH ARTICLE

Pharmacognostic, physicochemical, and phytochemical analysis of Sarasvata Churna - An antiepileptic Ayurvedic formulation

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Abstract

Background: Standardization of herbal medicines is essential to establish its identity, purity, quality, safety, and efficacy. This study reports on standardization of Sarasvata Churna an Ayurvedic formulation for long-term management of epilepsy and other mental disorders. **Objective:** This study aimed to prepare the Sarasvata Churna using authentic herbs and establishing pharmacognostical, physicochemical, and phytochemical standards for the formulation. **Materials and Methods:** Sarasvata Churna was prepared as per the formula and procedure mentioned in traditional texts and Ayurvedic Formulary of India. The prepared formulation was evaluated for pharmacognostical, physicochemical, and phytochemical parameters using guidelines of the World Health Organization and Pharmacopoeial Laboratory for Indian Medicines for quality control of herbal drugs. **Results:** Sarasvata Churna was obtained as a soft, fine, light brown, aromatic powder with salty bitter taste. Maximum extractive value of 38.67 ± 0.011% and minimum extractive value of 7.31 ± 0.01% were obtained in hydroalcoholic and petroleum ether, respectively. Ash values (total ash - 14.18 ± 0.005%, acid insoluble ash - 7.58 ± 0.055%, and water soluble ash - 4.42 ± 0.01%), foreign matter - 0.021 ± 0.02, loss on drying - 6.18 ± 0.01%, bitterness value - 2.6, swelling index - 2.17 ± 0.01, foaming index - below 100, and a volatile oil content - 2.40% were observed. No microbial load was detected in the formulation. Microscopic studies revealed the presence of various characteristic entities. Preliminary phytochemical screening confirms the presence of alkaloids, flavonoids, glycosides, fatty acids, terpenoids, sterols, tannins, proteins, and phenolic compounds. **Conclusion:** The various pharmacognostical, physicochemical, and phytochemical standards will help in quality control/quality assurance and maintaining batch to batch consistency in herbal drug industries so that maximum therapeutic efficacy can be achieved.

Key words: Pharmacognostic, physicochemical, phytochemical, Sarasvata Churna, Standardization, World Health Organization guidelines

INTRODUCTION

Polyherbal formulations, nowadays, are choices for the management of various diseases or disorders such as diabetes, hypertension, and hepatic diseases which requires therapy for the long duration of time. The use of herbal supplements has increased in the past decades due to high cost, decreased patient compliance, and associated toxicities of modern medicines. Epilepsy affecting more than 2% world population is one such neurological disorder which requires proper and continuous therapy to prevent or prolong the onset of seizures. Modern antiepileptic drugs such as hydantoin derivatives, iminostilbenes, valproates, barbiturates, succinimides, benzodiazepines, and amino acid derivatives have associated toxicities and degenerative effects on some essential biochemicals on prolonged use.

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Ayurvedic literature Sarangdhar Samhita has highlighted the concept of polyherbalism to achieve greater therapeutic efficacy. The active phytochemical constituents of individual herbs are insufficient to achieve the desirable therapeutic effects, but multiple herbs when combined in a particular ratio will give a better therapeutic effect and thus reduce the toxicity due to low concentrations of various phytochemicals.[1]

Sarasvata Churna is prescribed in Ayurveda for management of epilepsy and other mental disorders. Sarasvata Churna is not frequently prescribed by practitioners for the treatment and management of epilepsy due to non-availability of quality products in the market and lack of safety, efficacy, and clinical data. Maximum therapeutic benefits from a polyherbal formulation can be expected only when a quality preparation is used.

Sarasvata Churna as mentioned by Bhavaprakasha and Ayurvedic Formulary of India (AFI) is a compound preparation containing Kushta, Ashwagandha, Lavana, Ajamoda, Jeeraka, Krishna Jeeraka, Pippali, Maricha, Shunti, Patha, Shankapushpi, and Vacha powders triturated with Brahmi Swarasa. All the constituent herbs used in the formulation of Sarasvata churna possess pharmacological activities that either directly or indirectly help in the suppression of onset of seizures and thereby offer antiepileptic effects.[2]

Plant materials are used throughout developed and developing countries as home remedies, over-the-counter drug products, and raw materials for the pharmaceutical industry and represent a substantial proportion of the global drug market. The majority of adverse events reported in relation to the use of herbal products and herbal medicines are attributed to poor quality of the product. The World Health Organization (WHO) appreciated and stressed the use of standardized formulations and issued guidelines for establishing identity, purity, quality, and efficacy for herbal materials, within the overall context of quality assurance and control of herbal medicines.[3]

This study was aimed to prepare Sarasvata Churna as per the AFI using authentic herbs and establishing the quality control parameters. Physicochemical evaluation of component herb of the formulation, organoleptic, microscopic, phytochemical, and microbial load evaluations of Sarasvata Churna was carried out as per the procedures prescribed by the WHO, Geneva, and Pharmacopoeial Laboratory for Indian Medicines, Department of AYUSH, to set standards for use by herbal industries to avoid batch to batch variations and to maintain quality and consistency.[4]

MATERIALS AND METHODS

Plant Material

All the constituent herbs used in the formulation of Sarasvata Churna have been procured from Arya Vastu Bhandar, Dehradoon, and herbal garden of Ram-Eesh Institute of Vocational and Technical Education, Greater Noida. The herbs are authenticated by Dr. Sunita Garg, Emeritus Scientist of Raw Materials Herbarium and Museum Department (RHMD) of National Institute of Science Communication and Information Resources (NISCAIR), New Delhi. The authentication report for the plant materials *Piper longum* (fruits), *Piper nigrum* (fruits), *Saussurea lappa* C.B. Clarke (roots), *Withania somnifera* (L) Dunal (roots), Rock salt, *Acorus calamus* L. (rhizomes), *Convolvulus pluricaulis* Choisy (aerial part), *Bacopa monnieri* (L.) Penn.(whole plant), *Carum roxburghianum* Benth. Ex Kurz (fruit), *Carum carvi* L. (fruit), *Cuminum cyminum* L. (fruit), and *Zingiber officinale* Rosc. (rhizomes) has the reference no. NISCAIR/RHMD/Consult/2016/2993-20, whereas the authentication report for *Cissampelos pareira* L. (roots) has reference no. NISCAIR/RHMD/Consult/2016/3026-53. The voucher specimens of the samples are also preserved in the museum of Ram-Eesh Institute of Vocational and Technical Education, Greater Noida.

Formulation of Sarasvata Churna

Sarasvata Churna was prepared using finely powdered constituent herbs as per the formula given in the AFI.[5] The formula and plant part used of individual herbs are presented in Table 1.

All the constituent herbs are powdered individually and passed through Sieve No. 80. The fine individual powders

Table 1: Formula for preparing Sarasvata Churna

Plant part used	Name of herb	Quantity
Fruits	*P. longum* L.	1 part
Fruits	*P. nigrum* L.	1 part
Roots	*S. lappa* C.B. Clarke	1 part
Roots	*W. somnifera* (L.) Dunal	1 part
Rock salt	Sandha Namak, the Rock salt	1 part
Rhizomes	*A. calamus* L.	1 part
Aerial part	*C. pluricaulis* Choisy	1 part
Roots	*C. pareira* L.	1 part
Fruit	*C. roxburghianum* Benth. Ex Kurz	1 part
Fruit	*C. carvi* L.	1 part
Fruit	*C. cyminum* L.	1 part
Rhizome	*Z. officinalis* Rosc.	1 part
Whole plant Swarasa	*B. monnieri* (L.) Penn.	q.s for Bhavana

P. longum: Piper longum, P. nigrum: Piper nigrum, S. lappa: Saussurea lappa, W. somnifera: Withania somnifera, A. calamus: Acorus calamus, C. pareira: Cissampelos pareira, C. carvi: Carum carvi, C. cyminum: Cuminum cyminum, Z. officinalis: Zingiber officinalis, B. monnieri: Bacopa monnieri, C. pluricaulis: Convolvulus pluricaulis, C. roxburghianum: Carum roxburghianum

were weighed accurately and mixed together. Swarasa (Juice) of fresh Brahmi was used to give bhavana (lavigation) to the above powder mixture. Two Bhawanas were given to the powder, and slurry was then dried to form powder again which was then again sieved using Sieve No.80 #. The powder after drying and sieving was packed in an airtight container for further use. The procedure was well depicted in Figure 1.

Physicochemical Evaluations of Component Herbs[3,4,6-8]

The determination of various physicochemical parameters such as foreign matter, extractive values in different solvents, total ash, acid insoluble ash, water-soluble ash, moisture content or loss on drying, bitterness value, swelling index, foaming index, and pH value of 1% and 10% solutions was carried out by the methods given in the WHO guidelines for standardization of herbal drugs.

Macroscopic Evaluations

Sarasvata Churna was evaluated for organoleptic parameters such as color, taste, odor, touch, and texture. Powder fineness also determined using the procedure prescribed in the WHO guidelines for quality control of herbal drugs.

Microscopic Evaluation

Sarasvata Churna was evaluated for microscopic features using compound microscope at various magnifications. The powder was taken on slide mounted in glycerine and observed for specific characters. Staining with saffranine and iodine was also employed to get resolved images.[3]

Volatile Oil Content

Sarasvata Churna also evaluated for volatile content using hydrodistillation method using Clevenger assembly. 10 g Churna was used to obtain volatile content, and the percentage yield was calculated with reference to air-dried material used.[3]

Phytochemical Evaluations [8,9]

Sarasvata Churna was evaluated for the presence of reducing sugars using Fehling's test and Benedict's test, whereas Barfoed's test was used for monosaccharides. Cobalt chloride test was performed for the presence of hexose sugars. Solubility and filter paper stain test was used to detect the presence of fats and oils in the Sarasvata Churna. Amino acids were detected using Ninhydrin test, whereas biuret, xanthoproteic, 5% lead acetate, and 5% copper sulfate test were used for proteins. Alkaloids in the sample were screened using extracts of Sarasvata Churna in various solvents by

Figure 1: Various ingredients of Sarasvata Churna. Plate 1: *Carum roxburghianum*, Plate 2: *Saussurea lappa*, Plate 3: Rock salt, Plate 4: *Withania somnifera*, Plate 5: *Convolvulus pluricaulis*, Plate 6: *Piper longum*, Plate 7: *Piper nigrum*, Plate 8: *Cissampelos pareira*, Plate 9*: Acorus calamus*, Plate 10: *Carum carvi*, Plate 11: *Cuminum cyminum*, Plate 12: *Zingiber officinale,* Plate 13: *Bacopa monnieri,* Plate 14: Powder under Bhavana with Bacopa juice, Plate 15: Sarasvata churna after bhavana and drying

Dragendorff's, Hager's, Wagner's, and Mayer's test, whereas filter paper stain test was performed to detect terpenoids.

Salkowski test and Liebermann–Burchard tests were employed for the presence of sterols. Cardiac glycosides were detected using Keller–Killiani and Legal's test, whereas Borntrager's and modified Borntrager's tests were carried out for confirming the presence of anthraquinone glycosides. The presence of saponins in Sarasvata Churna was carried out using foam test. Shinoda test and lead acetate tests were performed for flavonoids detection. Lead acetate test, bromine water test, and ferric chloride tests were used for qualitative analysis of tannins and phenolic compounds.

All the reagents and chemicals used in above tests were prepared fresh using quality chemicals and ingredients.

Fluorescence Evaluations

1mg powder of Sarasvata Churna was taken on a glass slide and treated with various reagents, acids, and alkali solutions, and color of the powder before and after treatment was observed under ultraviolet light at visible light, short, and long wavelengths.[10]

Microbial Load Evaluation

Sarasvata Churna was screened for the presence of microbial load as per the WHO guidelines for quality control of herbal medicines. The sample was screened for total bacterial count, yeast and moulds, *Enterobacteria, Escherichia coli*, *Pseudomonas aeruginosa*, *Salmonella* species, and *Staphylococcus aureus* using the prescribed procedure.[11]

Statistical Analysis

All the physicochemical evaluations were made in triplets except foaming index and pH determinations, so the result was presented as mean ± standard deviation.

RESULTS

In-house polyherbal formulation Sarasvata Churna was prepared as per the AFI, and the list of ingredients and formula is given in Table 1. The extractive values of various ingredient herbs and Sarasvata Churna in various solvents are given in Table 2. The various physicochemical parameters such as total ash, acid insoluble ash, water-soluble ash, loss on drying, swelling index, foaming index, volatile oil content, bitterness values, and pH value of 1% and 10% aqueous solutions of Sarasvata Churna are presented in Table 3. The results of fluorescence analysis of Sarasvata Churna are given in Table 4, whereas the results of phytochemical analysis are given in Table 5.

Organoleptic evaluation and powder fitness of Sarasvata Churna are given in Table 6. Microscopic evaluation of a powder of Sarasvata Churna revealed the presence of characteristic features which are presented in Figure 2 Microbial load in the Sarasvata Churna was found to be within limits and presented in Table 7.

Table 2: Extractive values of component herbs of Sarasvata churna

Constituent herb	Extractive values in different solvents					
	PE	CF	EA	ME	A	HA
Z. officinalis	6.37±0.32	8.60±0.19	6.84±0.11	14.8±0.18	19.84±0.49	31.51±0.57
A. calamus	16.63±0.93	6.38±0.27	6.73±0.10	16.34±0.36	16.34±0.58	27.32±0.27
S. lappa	5.57±0.24	8.67±0.32	6.67±0.22	13.54±0.12	26.27±0.37	29.30±0.67
C. pareira	6.19±0.13	8.51±0.19	6.47±0.16	11.32±0.22	18.42±0.19	23.66±0.14
C. carvi	7.71±0.09	8.35±0.24	6.50±0.25	6.47±0.35	16.61±0.26	18.71±0.38
Carum roxburghianum	5.44±0.22	7.43±0.20	4.48±0.05	10.40±0.20	14.43±0.17	17.40±0.27
C. cyminum	4.27±0.08	6.41±0.30	6.37±0.15	4.48±0.13	5.72±0.23	13.49±0.36
W. somnifera	3.62±0.09	9.63±0.09	5.47±0.36	18.23±0.02	20.51±0.32	26.48±0.48
P. longum	8.14±0.10	10.37±0.15	6.67±0.03	16.57±0.075	14.68±0.15	22.38±0.10
P. nigrum	8.59±0.31	12.66±0.28	6.59±0.32	18.46±0.09	12.54±0.16	14.67±0.18
B. monneire	9.24±0.08	16.32±0.11	14.75±0.07	26.40±0.29	20.01±0.19	26.47±0.18
C. pluricaulis	2.63±0.09	7.84±0.22	5.15±0.10	8.29±0.03	11.21±0.13	12.38±0.24
Sarasvata Churna	7.31±0.01	9.68±0.02	10.63±0.005	27.36±0.015	38.15±0.26	38.67±0.011

PE: Petroleum ether, CF: Chloroform, EA: Ethylacetate, ME: Methanol, A: Water; HA: Hydroalcoholic, *P. longum: Piper longum, P. nigrum: Piper nigrum, S. lappa: Saussurea lappa, W. somnifera: Withania somnifera, A. calamus: Acorus calamus, C. pareira: Cissampelos pareira, C. carvi: Carum carvi, C. cyminum: Cuminum cyminum, Z. officinalis: Zingiber officinalis, B. monnieri: Bacopa monnieri, C. pluricaulis: Convolvulus pluricaulis, C. roxburghianum: Carum roxburghianum*

Table 3: Physicochemical parameters

Constituent herb	Extractive values in different solvents									
	TA	AIA	WSA	FM	LOD	SI	FI	pH (1/10%)	VOC	BV
Z. officinalis	8.62±0.02	2.16±0.04	2.8±0.02	0.28±0.01	2.23±0.03	3.63±0.04	<100	6.1/6.8	-	-
A. calamus	6.082±0.02	0.52±0.006	3.29±0.04	1.83±0.03	8.15±0.035	8.64±0.03	<100	6.9/8.4	-	-
S. lappa	4.06±0.03	1.24±0.017	3.10±0.015	0.84±0.02	3.48±0.14	0.66±0.043	<100	6.2/6.8	-	-
C. pareira	7.64±0.00	2.58±0.04	2.32±0.01	1.22±0.04	2.29±0.03	0.78±0.04	<100	6.3/6.6	-	-
C. carvi	6.47±0.025	1.27±0.005	2.73±0.02	0.60±0.02	6.60±0.015	1.63±0.03	<100	5.1/6.1	-	-
Carum roxburghianum	6.07±0.02	1.12±0.01	1.65±0.006	2.53±0.02	2.81±0.04	2.63±0.05	<100	6.3/7.0	-	-
C. cyminum	6.82±0.02	0.68±0.04	0.9±0.03	0.34±0.02	8.32±0.05	1.76±0.03	<100	5.1/6.2	-	-
W. somnifera	5.34±0.03	0.52±0.01	1.23±0.01	0.84±0.05	10.26±0.03	1.27±0.03	<100	6.3/7.1	-	-
P. longum	5.34±0.02	0.46±0.006	1.33±0.025	0.75±0.03	12.61±0.02	0.86±0.04	<100	5.1/5.4	-	-
P. nigrum	7.90±0.02	2.61±0.03	1.84±0.02	0.67±0.02	7.78±0.03	0.6±0.04	<100	5.0/5.5	-	-
B. monneire	10.40±0.05	1.41±0.04	2.75±0.06	1.86±0.03	2.13±0.03	1.27±0.03	<100	6.6/5.8	-	-
C. pluricaulis	7.22±0.02	2.84±0.03	2.3±0.04	0.67±0.04	1.21±0.04	1.67±0.04	<100	7.0/7.3	-	-
Sarasvata Churna	14.18±0.005	7.58±0.055	4.42±0.01	0.021±0.02	6.18±0.01	2.17±0.01	<100	5.3/6.7	2.40	2.46

TA: Total ash; AIA: Acid insoluble ash; WSA: Water-soluble ash; FM: Foreign matter; LOD: Loss on drying; FI: Foaming index; SI: Swelling index; VOC: Volatile oil content; BV: Bitterness value, *C. roxburghianum: Carum roxburghianum*, *P. longum: Piper longum, P. nigrum: Piper nigrum, S. lappa: Saussurea lappa, W. somnifera: Withania somnifera, A. calamus: Acorus calamus, C. pareira: Cissampelos pareira, C. carvi: Carum carvi, C. cyminum: Cuminum cyminum, Z. officinalis: Zingiber officinalis, B. monnieri: Bacopa monnieri, C. pluricaulis: Convolvulus pluricaulis, C. roxburghianum: Carum roxburghianum*

Table 4: Observations for fluorescence analysis of Sarasvata Churna

Reagent	Color observed		
	Visible	Short ultraviolet	Long ultraviolet
None	Light brown	Light brown	Brown
Distilled water	Brown	Greenish	Greenish brown
1 N NaOH in water	Light brown	Greenish brown	Black
1 N NaOH in ME	Yellowish brown	Greenish brown	Blackish brown
50% Nitric acid	Yellowish green	Dark green	Black
50% HCl	Light brown	Black	Light brown
Concentrated H2SO4	Dark brown	Yellowish brown	Greenish brown
Acetone	Light brown	Creamish green	Black
Conc. HCl	Light brown	Greenish brown	Dark brown
CF	Light brown	Light brown	Brown

CF: Chloroform, ME: Methanol

DISCUSSION

The Sarasvata Churna was prepared using the prescribed plant part of the authenticated herbs as per the standard procedure mentioned in the AFI and stored in an airtight container for further use.[5] The result from extractive values shows that the Churna was having a maximum extractive value of 38.67 ± 0.011% in hydroalcoholic and minimum extractive value of 7.31 ± 0.01% in petroleum ether, indicating a large number of phytoconstituents in the hydroalcoholic solvent systems. A high level of ash values such as total ash - 14.18 ± 0.005%, acid insoluble ash - 7.58 ± 0.055%, and water soluble ash - 4.42 ± 0.01% are due to the presence of rock salt as one of the ingredients in the formulation. As Churna was prepared from highly pure and refined herbs, the foreign matter content of Sarasvata Churna was found to be 0.021 ± 0.02. A low moisture level was observed in Sarasvata Churna as a loss on drying - 6.18 ± 0.01% which can also be due to the presence of high levels of aromatic components in the Churna.

As the Churna contains "trikatu," the three acrid drugs *Piper longum*, *Piper nigrum,* and *Zingiber officinale* known to increase the bioavailability of drugs, a bitterness value of 2.6 was observed in Sarasvata Churna, and due to this, the

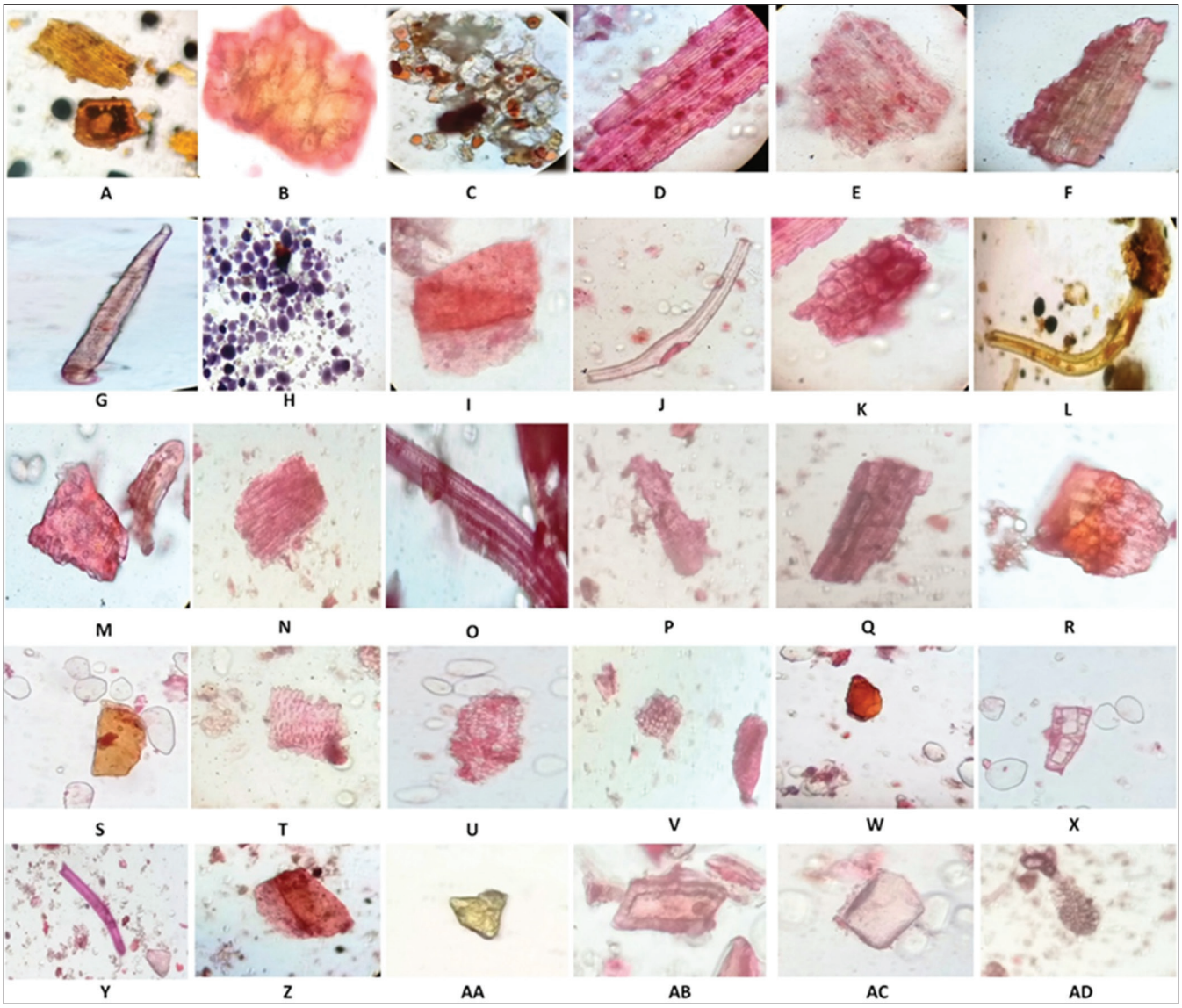

Figure 2: Powder microscopy of Sarasvata churna: A - Phloem fibers and schizogenous cell, B - Cork cells, C - Calcium oxalate crystals (intracellular and extracellular), D - Phloem fibers with cambium, E - Epidermal layer cells, F - Phloem fibers and parenchyma, G - Trichome, H - Starch grains, I - Epidermal cork cells, J - Phloem fibers, K- Epidermal cells, L - Trichome with parenchyma tissue, M - Epidermal cells with parenchyma, N - Xylem fibers, O - Vascular bundles, P - Sieve elements, Q - Annular xylem vessels, R - Cork cells, S - Resinous cell or Schizogenous cells, T - Schlerenchyma tissue, U - Parenchymatous tissues, V- Parenchymatous tissues, W- Oil granules, X - Quadrangular calcium oxalate crystals, Y - Phloem fibers, Z - Cork cells, AA - Crystal, AB - Stone cells, AC - Endosperm tissue, AD - Parenchyma with cork cells.

Churna must be administered with ghee or honey.[5] Due to the presence of resinous cells and high fiber content in the Churna, a swelling index of 2.17 ± 0.01 was observed. Phytochemical evaluations show the absence of saponins in the Churna which is well observed in a low foaming index of below 100.

Many component herbs of Sarasvata Churna contain volatile components which is also confirmed by the presence of a variety of oil globules and cells in the microscopic studies contribute to a high volatile oil content of 2.40%. Due to a high level of terpenoids, alkaloids, tannins, and other potential phytochemicals, the Churna itself possesses an antimicrobial profile which is confirmed by no fungal and bacterial load detected in the formulation.

Microscopic studies of Sarasvata Churna revealed the presence of various types and sized phloem fibers, schizogenous cell, cork cells, calcium oxalate crystals (intracellular and extracellular), cambium, epidermal layer cells, parenchymatous cells, trichomes, starch grains, xylem fibers, vascular bundles, sieve elements, sclerenchymatous tissues, oil granules, crystals, stone cells, and endosperm tissues which are characteristic entities of the constituent herbs of the formulation. Preliminary phytochemical screening confirms the presence of alkaloids, flavonoids,

Table 5: Phytochemical evaluation of Sarasvata Churna

Chemical tests	Extracts					
	Pet ether	Chloroform	Ethyl acetate	ME	Hydroalcoholic	Aqueous
Reducing sugars						
Fehling's test	-	-	-	+	+	+
Benedict's test	-	-	-	+	+	+
Monosaccharides						
Barfoed's test	-	-	-	-	-	-
Hexose sugars						
Cobalt–chloride test	-	+	+	+	+	+
Fats and oils						
Solubility test	+	+	+	+	+	-
Filter paper stain test	+	+	+	+	+	-
Amino acids						
Ninhydrin test	-	+	+	+	-	-
Proteins						
Biuret test	+	+	+	+	-	-
Xanthoproteic test	+	+	+	-	-	-
5% lead acetate test	+	+	+	-	-	-
5% copper sulfate test	-	+	+	+	-	-
Alkaloids						
Mayer's test	-	+	+	+	+	-
Hager's test	-	+	+	+	+	-
Wagner's test	-	+	+	+	+	-
Dragendorff's test	-	+	+	+	+	-
Terpenoids						
Filter paper stain test	-	+	-	+	+	-
Sterols						
Salkowski test	-	+	-	+	+	-
Liebermann–Burchard's test	-	+	+	+	+	-
Cardiac glycosides						
Keller–killiani test	-	-	-	-	-	-
Legal's test	-	+	-	-	-	-
Anthraquinone glycosides						
Borntrager's test	-	-	-	+	+	-
Modified Borntrager's test	-	-	-	+	+	-
Saponin glycosides						
Foam test	-	-	-	-	-	-
Flavonoids						
Shinoda test	-	-	-	+	+	-
Lead acetate test	-	-	-	+	+	-
Tannins and phenolics						
Lead acetate test	-	-	+	+	+	+
Bromine water test	-	-	+	+	+	+
Ferric chloride test	-	-	-	-	-	-

ME: Methanol

Table 6: Organoleptic characteristics and Powder fineness of *Sarasvata Churna*

S. No.	Parameters	Inference
1	Color	Light brown
2	Odor	Aromatic
3	Taste	Bitter salty with warmth sensation
4	Touch and Texture	Soft
5	Powder fineness	Sieve No. 80 (177µm) - 100%
		Sieve No. 85 (160 µm) - 21%
		Sieve No. 100 (150 µm) - 06%

Table 7: Microbial load estimation in *Sarasvata Churna*

S.No.	Parameter	Specifications	Observations	Inference
1.	Total Bacterial Count (TBC)	Medium used: Casein Soyabean Digest Agar Incubation temp.: 30–35°C. Incubation period: 5 Days. Dilution: 100 (1g in 100ml Lactose broath)	90 cfu/g (Dilution conc.: 0.001g/ml) 90X100X10=9.0X104	TBC within prescribed limits
2.	Yeast and Moulds	Medium used: Sabouraud Glucose Agar Incubation temp.: 20-25°C. Incubation period: 5 Days. Dilution: 10 (10g in 100ml Lactose broath)	No fungal growth observed	Nil.
3.	Enterobacteria	Medium used: Violet-red Bile Agar Incubation temp.: 35–37°C. Incubation period: 18–48 hours. Dilution: 10 (10g in 100ml Lactose broath)	No colonial growth	Nil
4.	*E. Coli*	Medium used: MacConkey agar Incubation temp.: 43–45°C. Incubation period: 18–24 hours. Dilution: 10 (10g in 100ml Lactose broath)	No red zones/colonies observed	Absent
5.	*Pseudomonas aeruginosa*	Medium used: Cetrimide Agar Incubation temp.: 35–37°C. Incubation period: 24–48 hours. Dilution: 10 (10g in 100ml Lactose broath)	No growth of microorganisms	Absent
6.	*Salmonella spp.*	Medium used: Brilliant Green Agar Incubation temp.: 35–37°C. Incubation period: 24–48 hours. Dilution: 10 (10g in 100ml Lactose broath)	No transparent/ colorless/opaque/pink/ white growth or zone	Absent
7.	*Staphylococcus aureus*	Medium used: Baird-Parker agar. Incubation temp.: 35–37°C. Incubation period: 24–48 hours. Dilution: 10 (10g in 100ml Lactose broath)	No Black colonies	Absent

glycosides, fatty acids, terpenoids, sterols, tannins, proteins, and phenolic compounds.

CONCLUSION

The various pharmacognostical, physicochemical, and phytochemical standards thus obtained from this study will help in establishing the identity, purity, quality, safety, and efficacy of Sarasvata Churna. These standards can be used by various industries and laboratories engaged in research and production of herbal formulations to control the quality of their products and help in maintaining batch to batch consistency so that maximum therapeutic efficacy can be achieved.

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REFERENCES

1. Parasuraman S, Thing GS, Dhanaraj SA. Polyherbal formulation: Concept of ayurveda. Pharmacogn Rev 2014;8:73-80.
2. Kaushik R, Jain J, Mazumdar A, Singh L. Studying the pharmacological basis of an antiepileptic Ayurvedic formulation-Sarasvata Churna. Int J Green Pharm 2017;11:1-7.
3. WHO, Geneva. Quality Control Methods for Mezdicinal Plant Materials. Updated edition. Geneva, Switzerland: WHO Press, World Health Organization; 2011. p. 1-187.
4. Lohar DR. Protocol for Testing Ayurvedic, Siddha and Unani Medicines. Ghaziabad: Pharmacopoeial Laboratory for Indian Medicines;2007. p. 1-146.
5. Ayurvedic Formulary of India. National Institute of Science Communication, CSIR. Part-II, First English Edition. New Delhi: Ayurvedic Formulary of India; 2000. p. 97-8.
6. Kaushik R, Sharma B, Jain J, Gupta D, Patel P. Establishment of monograph of *Acorus calamus* L. Rhizomes. J Drug Deliv Ther 2012;2:136-40.
7. Mukherjee PK. Quality Control of Herbal Drugs. 1st ed. India: Business Horizons Publishers; 2002. p. 195-6.
8. Khandelwal KR. Preliminary Phytochemical Screening. Practical Pharmacognosy. 19th ed. Pune: Nirali Prakashan; 2008. p. 149-56.
9. Hakim MA. Format for the Pharmacopoeial Analytical Standards of Compound Formulation, Workshop on Standardization of Unani Drugs, (appendix). New Delhi: Central Council for Research in Unani Medicine (CCRUM); 1995. p. 24-5.
10. WHO. WHO Guidelines for Assessing Quality of Herbal Medicines with Reference to Contaminants and Residues. Geneva, Switzerland: WHO Press, World Health Organization; 2007. p. 1-850.
11. Rai P, Rajput SJ. Preparation and physicochemical characterization of ingredients of Indian traditional medicine, Mahamrutyunjaya Rasa. J Ayurveda Integr Med 2017;8:159-68.

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