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Comparative Evaluation of In-House Prepared and Marketed Chaturbeeja Churna

Research Article

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Abstract

The standardization and quality control of herbs and herbal dosage forms with proper integration of modern scientific technique and traditional knowledge is important. The term 'standardization' refers to all actions undertaken during manufacturing and quality control to ensure consistent and reproducible product quality. The present study focuses on the comprehensive comparative evaluation of in-house prepared and marketed Chaturbeeja Churna. Chaturbeeja Churna is a classical ayurvedic formulation traditionally used for managing metabolic disorders. The formulation, comprises of four seeds viz. *Trigonella foenum-graecum* (Fenugreek), *Nigella sativa* (Kalonji), *Lipidium sativum* (Chandrasura), and *Trachyspermum ammi* (Ajwain). Each plant material were subjected to rigorous physicochemical, phytochemical, and quantitative estimation of secondary metabolites. These evaluation helps to assess quality, safety, and therapeutic potential of all the four seeds to be used in churna preparation. The in-house chaturbeeja churna was prepared according to the ayurvedic text. Standardized laboratory protocols were employed on Churna to determine parameters such as organoleptic, micromeritics, physicochemical and phytochemical properties. High-performance thin-layer chromatography (HPTLC) profiling was performed to evaluate chemical fingerprinting and ensure batch-to-batch consistency. The in-house formulation demonstrated superior physicochemical properties and a richer phytochemical profile, indicating better therapeutic efficacy. The marketed sample showed variability in certain physicochemical parameters, suggesting the need for stringent quality control practices. This study underscores the importance of standardization and quality assessment in the preparation of Ayurvedic formulations and provides a validated framework for future quality control of Chaturbeeja Churna and similar polyherbal products.

Keywords: *Chaturbeeja churna,* Ayurvedic formulation, *Methika, Chandrasura, Kalonji, Yavanika*.

Introduction

Chaturbeeja Churnam is a unique Ayurvedic formulation composed of four herbs that are Methika, Chandrashur, Kalonji and Yavanika mentioned in the ancient Ayurvedic text, *Bhavaprakasha Nighantu*. These herbs known for treating menstrual disorders, showcasing its curative properties in traditional medicine practices. It is primarily utilized for treating *Vataroga* (disorders related to the Vata dosha), as well as a range of conditions including *Ajeernam* (indigestion), *Soola* (pain), *Adhmanam* (abdominal distention), *Parshwashulam* (flank pain), and *Katishulam* (low back pain). (1)

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Evidence-Based Approach

The relevant literature for this study was sourced from the *Bhavaprakasha Nighantu* as well as databases like Google Scholar, PubMed, Web of Science, and the Ayush Portal. This evidence-based approach helps in understanding the formulation's effects in light of both classical and contemporary research.

Methika

Trigonella foenum-graecum, commonly known as fenugreek, belongs to the *Fabaceae* family and is cultivated extensively across India. While the name "Methika" may not be directly mentioned in the classical *Brihat Trayees*, fenugreek seeds have long been recognized in Ayurvedic medicine for their wide-ranging therapeutic properties. Traditionally, fenugreek has been used as a carminative, demulcent, expectorant, laxative, and stomachic agent.The seeds contain several bioactive compounds, including steroidal saponins such as diosgenin and gitogenin, essential oils, and proteins. These constituents contribute to its broad pharmacological effects, which include antioxidative, antineoplastic, anti-inflammatory, anti-ulcerogenic,

antipyretic, immunomodulatory, and anti-tumor properties.(2) Notably, fenugreek has been shown to interact with key proteins that activate the EGFR/AKT/mTOR signaling pathway, indicating its potential in managing hyperglycemia and hyperlipidemia.(3) Furthermore, polysaccharides like galactomannans have demonstrated anti-diabetic effects,(4) while the amino acid 4-hydroxy isoleucine possesses insulin-mimetic properties.(5)

Chandrasura

Lepidium sativum, or cress, is an annual herb belonging to the *Cruciferae* family. It is commonly used in Arab countries for treating respiratory conditions such as bronchitis and asthma.(6) The herb exhibits a variety of therapeutic properties, including antibacterial, aphrodisiac, diuretic, expectorant, gastrointestinal stimulant, gastroprotective, and laxative effects. Additionally, it is used for managing conditions like hemorrhoids, constipation, and swelling.(7) Chemical constituents like lepidine, imidazole, oleic acid, linoleic acid, and ascorbic acid contribute to its diverse pharmacological profile. Studies have demonstrated its antihypertensive, anti-inflammatory, analgesic, anticoagulant, antirheumatic, and hypoglycemic activities, among others. These findings support the traditional use of *Chandrasura* in a wide range of ailments.(8)

Kalonji

Nigella sativa, known as black cumin, is an annual herb from the *Ranunculaceae* family, renowned for its numerous therapeutic effects. The seeds are rich in essential oils containing compounds such as thymoquinone, cymine, nigellone, and carvone. Black cumin has been widely studied for its antimicrobial, anti-inflammatory, antioxidant, and immunomodulatory properties. It also possesses diuretic, antihypertensive, anticancer, and hepatoprotective activities. The therapeutic properties of *Kalajaji* are attributed largely to thymoquinone, a major active constituent, which has shown promise in the treatment of conditions such as asthma, bronchitis, rheumatism, gastrointestinal disorders, and skin diseases. Its wide range of effects also includes its use as an appetite stimulant, anti-diarrheal, and for managing parasitic infections.(9)

Yavanika

Trachyspermum ammi, or ajwain, belongs to the *Umbelliferae* family and is commonly used both as a culinary spice and for its medicinal properties. In Ayurveda, it is classified under *Sulaprasamanam*, which refers to remedies for alleviating pain, especially abdominal discomfort. *Yavanika* is known for its effectiveness in treating digestive issues such as indigestion, flatulence, and abdominal cramps.(10) This herb has a variety of therapeutic benefits, including bronchodilatory, antitussive, and antimicrobial properties. It is also used for its carminative effects, helping to relieve bloating and gas, and for treating gastrointestinal disorders like acid reflux, abdominal tumors, and infections caused by *Helicobacter pylori*. (11) Additionally, *Yavanika* has been shown to possess anti-carcinogenic, diuretic, and antidiarrheal effects.(12)

Comparison Between Modern Medicine and Ayurveda

The Ayurvedic and modern medical perspectives on the therapeutic uses of the herbs in *Chaturbeeja Churnam* align in many ways, though the mechanisms may be described differently. While modern science highlights the specific biochemical compounds and their effects on disease pathways, Ayurveda provides a broader framework that incorporates the balance of doshas and the qualities (guna) of each herb. Herbs like *Methika* are primarily used to manage *Kapha* disorders with additional effects on *Vata*, while *Chandrasura*, *Kalajaji*, and *Yavanika* are similarly effective in treating conditions associated with *Kapha* and *Vata* imbalances. (13-16)

This comparison between modern research and Ayurvedic principles opens the door for further exploration and integration of these healing systems. Therefore, in the present study the chaturbeeja churna was prepared in the laboratory as per the standard procedure from traditional literature and was evaluated simultaneously by comparing it with the marketed chaturbeeja churna. The evaluation of the sample was done by various analytical methods.

Materials and Methods

Procurement of plant materials and formulation:

The plant materials methika, chandrashur, kalonji and yavanika were purchased from Shivshankar Ayurvedic Pharmacy. All the plant materials were cleaned and dried properly. They were powdered using Wiley mill (HICON), passed through a #80 mesh sieve and stored separately in air tight containers. The marketed Chaturbeeja churna of brand Shri. Navjeevan Rasayanshala was purchased from amazon.

Figure 1: Plant Materials

Methika Chandrashur Kalonji Yuvanik MCC

The evaluation of drug means confirmation of its identity and determination of its quality and purity and detection of adulteration if any. The evaluation of each plant material is mandatory before using them in formulations. Therefore, as per WHO guidelines, the evaluation of quality and purity of plant materials has been carried out.(17)

Preliminary Phytochemical Screening (18)

The dried powder of each plant material was extracted with 70% ethanol at room temperature and evaporated under vacuum. The extract of plant material were labeled. All the extracts were subjected to preliminary phytochemical screening for testing various phytoconstituents such as carbohydrate, tannins, alkaloids, proteins, sterols, amino acids etc.

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Quantitative estimation of Phytoconstituents (19)

The quantitative estimation is carried out for amount determination of secondary metabolites in samples. The various estimations such as total alkaloid content, total flavonoid content and total phenol content were performed on selected plant materials extract by the standard procedure. (19) Estimation of Total alkaloids content by using Bromocresol Green reagent, Total phenolic content by Folin- Ciocalteu reagent, Total flavonoid content by Aluminium Chloride method and Total Saponin content were performed by n-butanol method.

Preparation and Evaluation of Chaturbeeja churna

The Chaturbeeja churna was prepared in laboratory by using the plant materials which were evaluated for assessing its quality and purity.

Preparation of Chaturbeeja Churna

Each ingredient were accurately weighed (10g). These powders were separately passed through sieve # 80 to prepare churna and then mixed together in 1:1:1 proportion. The prepared churna was labelled as LCC and evaluated for organoleptic characters, and micromeritic properties.

Table 1: Composition of Chaturbeeja Churna

Sr. No.	Ingredients	Quantity (g)
1	**Methika Seeds -MS**	10g
2	**Chandrashur Seeds - CS**	10g
3	**Kalonji Seeds - KS**	10g
4	**Yavanika Seeds - YS**	10g

The in-house Chaturbeeja Churna (LCC) was prepared which is shown in fig no 2 a. and the marketed Chaturbeeja Churna (MCC) in fig 2 b. and were selected for the further study.

Figure 2: In-house Churna and marketed Churna

a. In- house Chaturbeeja Churna (LCC)

b. Marketed Chaturbeeja Churna (MCC)

Organoleptic evaluations

The organoleptic evaluation of LCC and MCC was conducted for the characteristics such as color, odour, taste, texture.

Micromertic Evaluation:

Both the formulations, LCC and MCC were evaluated for the micromeritic properties such as, Bulk density, Tapped density, Angle of repose, Carr's index and Hausner's ratio. (18)

Physicochemical Evaluation

LCC and MCC were evaluated for their physicochemical properties such as, total ash value, acid in soluble ash value, water soluble ash value, water soluble extractive value and loss on drying.**(17)**

Preliminary Phytochemical screening:(19)

The various chemical tests were performed on LCC and MCC for the presence of various secondary metabolites such as carbohydrates, proteins, tannins, alkaloids, saponin, flavonoids, steroids, etc.

High Performance Thin Layer Chromatography (21)

The methanolic extracts of both LCC and MCC were subjected to High Performance Thin Layer Chromatographic (HPTLC) studies, to determine the probable number of compounds present. The standard procedure for developing the chromatographic profile of the sample was followed. The chromatographic fingerprinting of MCC and LCC formulations were done by using HPTLC method. Camag HPTLC system comprising of CAMAG Nanomat 4 sample applicator and TLC scanner 4 controlled by win CATS software version 3.1.5 was used for HPTLC method. Stationary phase used was MERCK precoated TLC Aluminium foil silica gel 60 F254 and the mobile phase Toluene: ethyl acetate: formic acid (6:4:0.3). Samples LCC and MCC were applied as spots with 10mm distance between two tracks. Tank saturation and plate equilibrium was given with filter paper for 20 min. Ascending development for a distance of 80 mm in a twin trough chamber was completed in 15 min. Samples i.e MCC and LCC was optimized for fingerprinting.

Observations and Results

Physicochemical Evaluation of crude drugs:

The results of all the physicochemical evaluation method are reported in table no.2.

Table 2: Results of Physicochemical evaluation of plants materials- MS, CS, KS, YS

Sr. No	Parameters	Methika - MS	Chandrashur - CS	Kalonji -KS	Yavani -YS
1	**Total ash value**	3.9%	2.5%	41%	5.6%
2	**Acid insoluble ash**	0.8 %	0.6%	1.9%	0.45%
3	**Water soluble ash**	2.73%	0.82%	2.1%	3.2%
4	**Petroleum soluble extractive value**	11.2%	12.5%	19.2%	17.4%
5	**Water soluble extractive value**	7.2%	9.4%	8.1%	21%
6	**Loss on drying**	0.9%	5%	1.95%	2.4%

All the physicochemical parameters determined for plant materials (MS, CS, KS, YS), complies the standard findings reported in various official literature. These findings suggest that all the four plant materials selected and evaluated were of good quality.

Preliminary Phytochemical Screening

The results of all the Preliminary Phytochemical Screening are reported in table no.3.

Table 3: Results of Preliminary phytochemical screening of plant materials

S. N.	Chemical test	Methika - MS	Chandrashur - CS	Kalonji -KS	Yavani -YS
1	Carbohydrates	+	+	+	+
2	Proteins	+	+	+	+
3	Tannins	-	-	+	-
4	Alkaloids	+	+	+	+
5	Saponins	+	-	+	+
6	Flavonoids	+	+	+	+
7	Steroids	-	-	-	-

Where, (-) represents absences of phytoconstituents, (+) represents presences of phytoconstituents.

Thus, table 3 reveals that MS contain carbohydrates, alkaloids, saponin and flavonoids. CS contains carbohydrates, alkaloids and flavonoids. KS contains carbohydrates, proteins, tannins, alkaloids, saponin and flavonoids. YS contains carbohydrates, proteins, alkaloids, saponins and flavonoids.

Quantitative Estimation of Phytoconstituents:

The results of all the Quantitative estimation are reported in table no.4.

Table 4: Quantitative estimations of plant extract

SN	Plant Extract	Total Alkaloid Content (mg atropine Equivalent /g extract)	Total Phenolic Content (mg Gallic Acid equivalent / g extract)	Total Flavanoid Content (µg/mg of quercetin /g extract)	Total Saponin Content (%)
1	MS	0.67	77.44	38.34	18-21%
2	CS	1.17	121.86	44.61	-
3	KS	0.35	98.84	42.65	5-7%
4	YS	0.65	76.05	39.61	1-2%

Thus, the table no. 4 reveals that the total alkaloid present in extracts of MS, CS, KS and YS found to be 0.6mg/g, 1.17mg/g, 0.35mg/g and 0.65mg/g respectively. The total polyphenol content in MS extract was found to be 77.44µg/mg, CS extract was found to be 121.86µg/mg, in KS extract it was found to be 98.84 µg/mg and in YS extract it was found to be 76.05µg/mg**.** The total flavonoid content in MS extract was found to be 38.34. µg/mg, in CS extract it was found to be 44.61 µg/mg and in KS extract it was found to be 42.65 µg/mg and total flavonoid content in YS extract was found to be 39.61 µg/mg, The total flavonoid content of CS was significantly higher than that of the other extracts i.e. CS, KS and YS. The total saponin content in MS was found to be 18-21 %, in KS it was found to be 5-7% and in YS it was found to be 1-2%.

After evaluating the plant extracts the formulation was prepared. This evaluation is important for standardization and assuring the quality of plant materials. The formulation was further studied for its organoleptic evaluations, micromeritic properties, physicochemical evaluations, phytochemical screening, and chromatographic fingerprinting by TLC and HPTLC.

Organoleptic Evaluations

Organoleptic evaluation of both marketed and in-house chaturbeeja churna shows that colour of LCC shows light brown and MCC shows dark brown, odour of LCC is aromatic and characteristic while odour of MCC is characteristic, taste of both LCC and MCC was bitter in taste and texture of LCC was fine and MCC shows coarse texture.

Table 5: Organoleptic Evaluation

S.N	Observations	LCC	MCC
1	Colour	Light brown	Dark brown
2	Odour	Aromatic and characteristic	Characteristics
3	Taste	Bitter	Bitter
4	Texture	Fine	coarse

Micromeritics Evaluation

For evaluating the flow properties of the powdered plant material selected in this study the parameters such as Bulk density, Tapped density, Carr's index, Angle of repose, Hausner's ratio were performed on in- house as well as marketed chaturbeeja churna.

Table no. 6: Micromeritics evaluation

Sr No.	Parameters	LCC	MCC
1	Bulk density	0.33 gm/cm^3	0.49 gm/cm^3
2	Tapped density	0.49 gm/cm^3	0.66 gm/cm^3
3	Carr's index	32.6 %	25.75%
4	Hausner's ratio	1.48	1.34
5	Angle of repose	25.20◦	22.47◦

The results depicted in Table no. 6 reveals that there was no significant difference between results of in house as well as marketed churna. Bulk density of chaturbeej churna was found to be 0.33 gm/cm^3 (F1), 0.49 gm/cm^3 (F2). Tapped density was found to be 0.49 (F1), 0.66 gm/cm^3 (F2). Carr's index was found to be 32.6 % (F1), 25.75 % (F2). Hausner's ratio was found to be 1.48, 1.34 and Angle of repose was found to be 25.20◦, 22.47◦. From this values both powders we understand chaturbeeja churna powder possess good flow properties.

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Physicochemical Evaluation

The results of Physicochemical evaluations are depicted in table no. 7.

Table no. 7 Physicochemical evaluations

Sr. No	Parameters	LCC	MCC
1	Total ash value	0.16g	0.19g
2	Acid insoluble ash	0.6g	0.8g
3	Water soluble ash	0.90g	0.95g
4	Water soluble extractive value	2.63g	2.60g
5	Loss on drying	1.4g	1.8g

The results of physicochemical evaluation of chaturbeeja churna are repored in Table no.7. based on the result, it was observed that the total ash value of marketed chaturbeeja churna was slightly more than laboratory chaturbeeja churna. Almost every parameter shows that the LCC was found to be of better quality.

Preliminary Phytochemical Screening

The result of chemical evaluation of in-house LCC and marketed *chaturbeej churna* MCC are reported in table no. 8.

Table 8: Preliminary phytochemical Screening of

Sr No.	Chemical	LCC	MCC
1	Carbohydrat	+	+
2	Proteins	+	+
3	Tannins	-	-
4	Alkaloids	+	+
5	Saponins	+	+
6	Flavonoids	+	+

Thus, the results of table no. 8 reveals that the Chaturbeeja formulations shows presence of Carbohydrates, Proteins, Alkaloids, Saponins and Flavonoids.

Chromatographic fingerprinting of LCC and MCC

The chromatographic fingerprints of both LCC and MCC obtained from HPTLC method are shown in fig 3. The distinct spots were observed on track 1 and 2 . The samples had shown much similarity in peaks under UV 364 scanner.

The HPTLC results indicate that 13 peaks were detected on track 1 and 12 peaks were detected on track 2 under the specified conditions. The Rf values and Area in % are presented in figure 5 and 6. This suggests that these particular components are common among the Methanolic extract of LCC and MCC. Thus, it can be concluded that the same compounds are present in the samples LCC and MCC on track 1 and 2 respectively.

Figure no. 3. Chromatogram of LCC and MCC on track 1 and 2 respectively in solvent system B under UV 364.

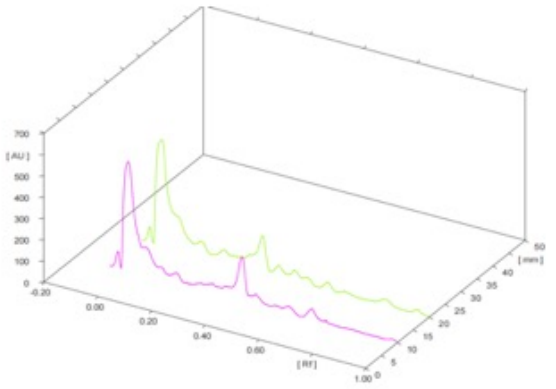
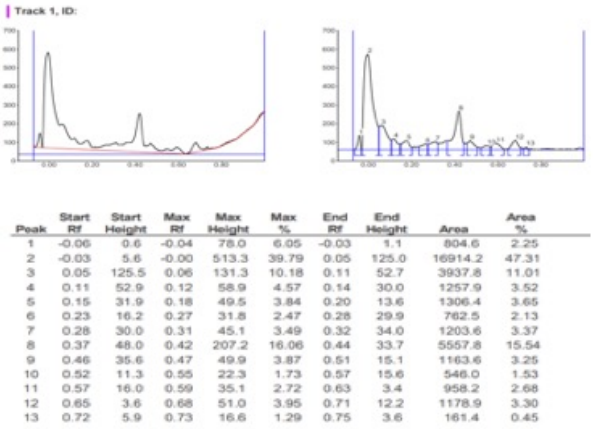
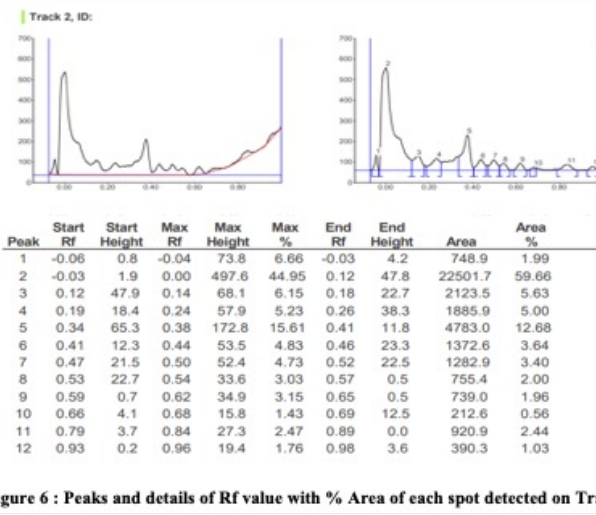


Figure no. 4. 3D representation of HPTLC of LCC and MCC



Peak	Start Rf	Start Height	Max Rf	Max Height	Max %	End Rf	End Height	Area	Area %
1	-0.06	0.6	-0.04	78.0	6.05	-0.03	1.1	804.6	2.25
2	-0.03	5.6	-0.00	513.3	39.79	0.05	125.0	16914.2	47.31
3	0.05	125.5	0.06	131.3	10.18	0.11	52.7	3937.8	11.01
4	0.11	52.9	0.12	58.9	4.57	0.14	30.0	1257.9	3.52
5	0.15	31.9	0.18	49.5	3.84	0.20	13.6	1306.4	3.65
6	0.23	16.2	0.27	31.8	2.47	0.28	29.9	762.5	2.13
7	0.28	30.0	0.31	45.1	3.49	0.32	34.0	1203.6	3.37
8	0.37	48.0	0.42	207.2	16.06	0.44	33.7	5557.8	15.54
9	0.46	35.6	0.47	49.9	3.87	0.51	15.1	1163.6	3.25
10	0.52	11.3	0.55	22.3	1.73	0.57	15.6	546.0	1.53
11	0.57	16.0	0.59	35.1	2.72	0.63	3.4	958.2	2.68
12	0.65	3.6	0.68	51.0	3.95	0.71	12.2	1178.9	3.30
13	0.72	5.9	0.73	16.6	1.29	0.75	3.6	161.4	0.45

Figure 5 : Peaks and details of Rf value with % Area of each spot detected on Track 1



Peak	Start Rf	Start Height	Max Rf	Max Height	Max %	End Rf	End Height	Area	Area %
1	-0.06	0.8	-0.04	73.8	6.66	-0.03	4.2	748.9	1.99
2	-0.03	1.9	0.00	497.6	44.95	0.12	47.8	22501.7	59.66
3	0.12	47.9	0.14	68.1	6.15	0.18	22.7	2123.5	5.63
4	0.19	18.4	0.24	57.9	5.23	0.26	38.3	1885.9	5.00
5	0.34	65.3	0.38	172.8	15.61	0.41	11.8	4783.0	12.68
6	0.41	12.3	0.44	53.5	4.83	0.46	23.3	1372.6	3.64
7	0.47	21.5	0.50	52.4	4.73	0.52	22.5	1282.9	3.40
8	0.53	22.7	0.54	33.6	3.03	0.57	0.5	755.4	2.00
9	0.59	0.7	0.62	34.9	3.15	0.65	0.5	739.0	1.96
10	0.66	4.1	0.68	15.8	1.43	0.69	12.5	212.6	0.56
11	0.79	3.7	0.84	27.3	2.47	0.89	0.0	920.9	2.44
12	0.93	0.2	0.96	19.4	1.76	0.98	3.6	390.3	1.03

Figure 6 : Peaks and details of Rf value with % Area of each spot detected on Track 2

Discussion

The standardization and quality control of herbal dosage form with proper integration of modern scientific technique and traditional knowledge is important. The route methods of herbal drug standardization addresses quality related issues using several evaluation parameters of crude drugs and chromatographic profiling assisted characterization with spectroscopic techniques. In case of the herbal dosage form the validated assays of the content of constituents are required along with details of analytical procedure.

The assays of marker substances or other justified determinations are required.

The present investigation involves evaluation of the plant materials used to prepare formulations, preparation of churna and evaluation of all the prepared as well as marketed chaturbeeja formulations.

All the physicochemical parameters determined for the plant materials (MS, CS, KS, YS), complies the standard findings reported in various official literature. Thus the findings suggest that all the four plant material selected and evaluated were of good quality. The preliminary phytochemical screening reveals that MS contain carbohydrates, alkaloids, saponin and flavonoids. CS contains carbohydrates, alkaloids and flavonoids. KS contains carbohydrates, proteins, tannins, alkaloids, saponin and flavonoids. YS contains carbohydrates, proteins, alkaloids, saponins and flavonoids.

These evaluated crude drug materials were used to prepare LCC. Organoleptic evaluation of both marketed (MCC) and in-house chaturbeeja churna (LCC) shows that colour of LCC was light brown and MCC was dark brown, odour of LCC is aromatic and characteristic odour of MCC is characteristic, taste of both LCC and MCC was bitter in taste and texture of LCC was fine and MCC was found to be course as compared to LCC. The flow properties such as Bulk density, Tapped density, Carr's index, Angle of repose, Hausner's ratio were performed for in house as well as marketed churna. There was no significant difference between results of LCC well as MCC. Bulk density of chaturbeej churna was found. The preliminary phytochemical screening reveals that the both chaturbeeja formulations LCC and MCC mainly contains carbohydrates, Protein, Tannin, Alkaloids, Saponins and Flavonoids. The chromatographic fingerprinting of the churna samples were obtained by HPTLC. The results indicate that each sample, LCC and MCC on track 1 and 2 share compounds with specific RF value, suggesting a common components in these samples. This suggests that these particular components are common among the Methanolic extract of LCC and MCC. Thus, This HPTLC method can be further utilized by the researchers for standardization of the chaturbeeja churna.

Conclusion

Standardization of *chaturbeeja churna* formulations was done using pharmacognostical and physicochemical parameters, chemical evaluation, HPTLC fingerprinting and proximate analysis of active constituents. Marketed sample was also evaluated and compared with in- house formulation. There was variation between marketed and in house formulations regarding ash values, extractive values and total phenolics. These variations may be due to change in the quality of raw materials. It is generally realized that for monitoring quality the standardization is needed which is performed here. Hence, the results of these compound can be kept as a standard for comparison and evaluation of other commercial samples of chaturbeeja churna available in the market. The method was found to be useful in detecting the genuineness of the herbal formulations.

Future scope

In future the Quantitative estimation by HPTLC can be done by using various markers.

References

1. Bhavamishra. *Bhavaprakasha Nighantu* (K. C. Chunekar, Ed.; G. S. Pandey, Comm.). Haritakyadi Varga, verse 23. Varanasi; Chaukhambha Bharati Academy; 2004.
2. Krishnasamy V, Patel A. Pharmacological Intervention of Classical Ayurvedic Formulation Chaturbeeja churnam – A Narrative Review Based on Guna Concept. *Pharmacog Rev.* 2021;15(30):186-90.
3. Yadav Umesh CS, Baquer Najma Z. Pharmacological effects of *Trigonella foenum-graecum* L. in health and disease. *Pharm Biol.* 2014;52(2):243-54.
4. Banerjee Subhadip, Bhattacharjee Pritorthi, Kar Amit, Mukherjee Pulok K. LC-MS/MS analysis and network pharmacology of *Trigonella foenum-graecum* – A plant from Ayurveda against hyperlipidemia and hyperglycemia with combination synergy. *Phytomedicine.* 2019;60:152944.
5. Madar Z, Shomer I. Polysaccharide composition of a gel fraction derived from fenugreek and its effect on starch digestion and bile acid absorption in rats. *J Agric Food Chem.* 1990;38(7):1535-39.
6. Broca Christophe, Breil Vincent, Cruciani-Guglielmacci Céline, Manteghetti Michèle, Rouault Christine, Derouet Michel, Rizkalla Salwa, Pau Bernard, Petit Pierre, Ribes Gérard, Ktorza Alain, Gross René, Reach Gérard, Taouis Mohammed. Insulinotropic agent ID-1101 (4-hydroxyisoleucine) activates insulin signaling in rat. *Am J Physiol Endocrinol Metab.* 2004;287(3):E463-71.
7. Baquar SR. *Medicinal and poisonous plants of Pakistan*. Karachi; Printas; 1989.
8. Duke JA, Bogenschutz-Godwin MJ, Ducelliar J, Duke PAK. *Handbook of medicinal herbs*. Boca Raton, FL; CRC Press; 2002.
9. Ahmad Aftab, Husain Asif, Mujeeb Mohd, Khan Shah Alam, Najmi Abul Kalam, Siddique Nasir Ali, Damanhouri Zoheir A, Anwar Firoz. A review on therapeutic potential of *Nigella sativa*: A miracle herb. *Asian Pac J Trop Biomed.* 2013;3(5):337-52.
10. Sharma PV. *Samhita Caraka (Text with English Translation)*. Reprint ed. Varanasi; Chaukhambha Orientalia; 2011. Volume 1, p. 29.
11. Boskabady Mohammad Hossein, Alitaneh Saeed, Alavinezhad Azam. *Carum copticum* L.: a herbal medicine with various pharmacological effects. *BioMed Res Int.* 2014;2014:569087.
12. Hanif Muhammad Awais, Hassan Syeda Mona, Mughal Shahzad Sharif, Rehman Aesha, Hassan Syed Khurram, Ibrahim Asif, Hassan Huma. An overview on ajwain (*Trachyspermum ammi*)

pharmacological effects: current and conventional. *Pharm Sci Technol.* 2021;5(1):1-6.

13. Sastry Dr. JLN. *Vijnana Dravyaguna*. Reprint ed. Varanasi; Chaukhambha Orientalia; 2010. p. 728-9.
14. Sastry Dr. JLN. *Vijnana Dravyaguna*. Reprint ed. Varanasi; Chaukhambha Orientalia; 2010. p. 598-9.
15. Sastry Dr. JLN. *Vijnana Dravyaguna*. Reprint ed. Varanasi; Chaukhambha Orientalia; 2010. p. 570-1.
16. Sastry Dr. JLN. *Vijnana Dravyaguna*. Reprint ed. Varanasi; Chaukhambha Orientalia; 2010. p. 269-70.
17. *Indian Pharmacopoeia*. 8th ed. Volumes I and II. New Delhi; The Controller of Publication, Ministry of Health and Family Welfare, India; 2018.
18. Khandelwal K.R. *Practical Pharmacognosy: Techniques and Experiments*. 8th ed. Pune; Nirali Prakashan; 2008. p. 149-156.
19. Khadabadi S.S. *Textbook of Pharmacognosy and Phytochemistry: A Comprehensive Approach*. 2nd ed. New Delhi; Nirali Prakashan; 2024. p. 129-132.
20. Subhramanyam C.V.S. *Textbook of Physical Pharmacy*. 2nd ed. Reprint 2010. New Delhi; Vallabh Prakashan; 2000. p. 180-234.
21. Meena AK, Narasimhaj CV, Velvizhi D, Singh A, Rekh P, Kumar V, Ilavarasan R, Narayanam S, Dhiman KS. Determination of Gallic Acid in Ayurvedic Polyherbal Formulation *Triphala churna* and its ingredients by HPLC and HPTLC. *Res J Pharm Technol.* 2018;11(8):3243-49.

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