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COMPARATIVE STANDARDIZATION STUDY OF GANDHARVA HARITAKI CHURNA FORMULATION

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ABSTRACT: Standardization is the need of the hour in Ayurvedic system of medicine. The traditional systems of medicine are really effective but the problem with them is they lack in quality assurance. This enables us to recognise the quality of the formulation. The Central Council of Research in Ayurveda and Siddha has prescribed the preliminary guidelines for testing the quality of these formulations. It is essential to derive a protocol or develop methods for evaluation of herbal formulation to maintain uniformity between batches during production. The present work aims to standardize a poly-herbal churna called Gandharva haritaki Churna available in the market. The churna was procured and standardised for the parameters like organoleptic characters, physical characters, physiochemical properties and phytochemical screening *etc*. These parameters can determine the quality of the product. The results were found to be within the standards.

INTRODUCTION:

Introduction of the Sample: Ayurvedic science has got its rich heritage in India. People in India believe that natural products are safe compared to synthetic drugs. The development in these traditional systems of medicine leads to maintain proper quality of the product. India is rich in its flora and fauna [1]. These plants are being used for curing many diseases as such in raw condition rather the being prepared as formulation; standardisation is an essential parameter to be done. It is a vital step in formulation since it determines the quality of the product and is essential to develop a protocol on standardisation of every product available in the market to avoid variation arising between batch to batch [2].

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FIG. 1: SAMPLE NO. 1 GANDHARVA HARITAKI CHURNA

Plant materials are not like synthetic drugs, they vary in many conditions even in their chemical content depending on the time and season of collection of plant material, the geographical location of the plant being grown *etc*. [3]

The CCRAS and WHO has introduced certain standards and guidelines to maintain uniformity between the production batches. Good manufacturing practices and quality control of the ingredients and products can result in ensuring quality assurance of the formulation [4].

FIG. 2: SAMPLE NO. 2 LAB MADE GANDHARVA HARITAKI CHURNA

The present study is to standardise a poly-herbal formulation available in the market called as Gandharva haritaki Churna used to treat many ailments of the body. The churna is evaluated for organoleptic properties, physical properties, physiochemical parameters and phytochemical screening to standardise the same.

MATERIALS AND METHODS: Gandharva haritaki Churna was selected because it had no previous specific scientific works been reported. So to prepare the standardisation procedures of the churna [5, 6], the present work was attempted. Gandharva haritaki Churna was procured from local market.

Organoleptic Evaluation: The colour, odour and taste of the formulation were evaluated manually **Table 1**.

TABLE 1: ORGANOLEPTIC ANALYSIS

S. no.	Parameters	Observation for Marketed Churna	Observation for Lab Made Churna
1	Odour	Characteristic	Characteristics
2	Taste	Astringent	Astringent
3	Colour	Light Yellowish	Light Yellowish
4	Texture	Fine	Fine

Physico-chemical Parameters: Loss on drying, ash values, extracting values, pH and crude fibre content [7] was determined for the physicochemical parameters **Table 2**.

TABLE 2: PHYSICOCHEMICAL EVALUATION

S. no.	Parameters	Observation for Marketed Churna	Observation for Lab made Churna
1	LOD (%)	1.8	2.5
2	pH 1% & 10% w/w	6 (acidic)	6 (acidic)
3	Total ash value (% w/w)	6.35	7.57
4	Acid insoluble ash value (% w/w)	3.4	3.18
5	Water soluble ash value (% w/w)	3.5	3.58
6	Crude fibre content (g)	4.2	3.5
7	Alcohol soluble extractive value (% w/w)	1.52	1.2
8	Water soluble extractive value(% w/w)	3.24	3.0

Loss on Drying: 2g of the churna was accurately weighed and transferred into a pre-weighed watch glass [8]. This was dried at 105 °C for 5 hrs with regular check of weight for every interval. The final loss in weight was calculated by

$$LOD\ (\%) = initial - final/initial \times 100$$

pH: pH of the churna was determined using pH meter by dispersing 1% w/v and 10% w/v churna in water.

Crude Fibre Content: 2g of the churna wad added with 50ml of 10% nitric acid. This was boiled and filtered. The retains was washed with hot water and added with 50ml of 2.5% v/v sodium hydroxide solution. This was again filtered, washed with hot water and the residue was transferred into a crucible. The weight of the residue was taken for determining the crude fibre present in the churna.

Ash Value: Total Ash Value: The total ash content was determined by taking 2g of churna into a pre-weighed and tarred crucible and incinerated at a temperature not exceeding 450 °C, cooled and weighed. The difference between initial and final gives the total ash value [9].

Acid Insoluble Ash: The residue of ash obtained in total ash was added with 25 ml of dilute HCl and boiled for 5 min. This was filtered using ashless filter paper and ignited again to determine the acid insoluble ash.

Water Soluble Ash Value: The residue of the total ash was added with 25 ml of water in the place of dil. HCl and the procedure was followed the similar way.

Extractive Value: [10]

Alcohol Soluble Extractive Value: 5g of churna was added with 100ml of alcohol and kept for 24 hrs, occasionally shaking and left aside after the first 6 h. It was then filtered. The filtrate was evaporated until constant weight was obtained. The difference in weight gives alcohol soluble extractive value.

Water Soluble Extractive Value: [11] 5g of churna was added with 100ml of chloroform water and kept for 24 h and the similar procedure was followed like alcohol soluble extractive value.

Physical Characteristics of Churna: [12] Bulk density, Angle of repose, Hausner's ratio, Carr's index, and particle size distribution was determined for evaluating the physical characteristics of the churna **Table 3**.

TABLE 3: PHYSICAL EVALUATION

S. no.	Parameters	Observation for marketed Churna	Observation for Lab made Churna
1	Bulk density (g/ml)	0.555	0.55
2	Tapped density (g/ml)	0.80	0.71
3	Angle of Repose (θ°)	35.75	33.42
4	Compressibility index (%)	30.625	22.53
5	Hausner's ratio	1.44	0.16

Bulk Density: 10 g of churna was taken in a graduated measuring cylinder and tapped on a wooden surface. Bulk density is calculated by using the formula.

$$\text{Bulk density} = \text{weight taken} / \text{bulk volume}$$

$$\text{Tap density} = \text{weight of churna taken} / \text{volume (tapped)}$$

Angle of Repose: Angle of repose was determined by using funnel method. The powder was allowed to flow through a funnel fixed on a stand to form a heap. The height and the radius give the angle of repose.

$$\text{Angle of repose } \tan\theta = h / r$$

$$\theta = \tan^{-1}(h/r)$$

Where, h = height of heap; r = radius of heap.

Compressibility / Carr's Index: This is calculated using the formula:

$$\text{Compressibility / Carr's Index} = \text{Bulk density (Tapped)} - \text{Bulk density (Untapped)} / \text{Bulk density (Tapped)} \times 100$$

Hausner's Ratio: The formula used to determine Hausner's ratio is

$$\text{Hausner's ratio} = \text{Bulk density (Tapped)} / \text{Bulk density (untapped)}$$

Fluorescence Analysis: A little amount of churna was macerated with a small quantity of solvents like 1N Sulphuric acid, 1N Nitric acid, 1N Hydrochloric acid, Iodine, Potassium hydroxide, Ammonia, 1N Sodium hydroxide for an hour and then filtered.

The filtrate was then analysed under day light and UV light for colour and fluorescence [13] **Table 4** and **5**.

Qualitative Phytochemical Screening: [15-17]

Detection of Tannins: 2-3 ml of aqueous or alcoholic extract of powders were tested carefully with various tannins test reagents as

- **5% $FeCl_3$ Solution:** A deep blue-black colour indicates the test is positive.
- **Lead Acetate Solution:** A white precipitate indicates the test is positive.
- **Bromine Water:** Deceleration of bromine water indicates the test is positive.

Dilute Iodine Solution: Transient red colour indicates the test is positive.

TABLE 4: FLUORESCENCE ANALYSIS FOR SAMPLE 1

Solvent added	Colour observed under		
	Day light	Short UV wavelength (256 nm)	Long UV wavelength (365 nm)
1N Sulphuric acid	Light brown	Light green	Dark Green
1N Nitric acid	Light brown	Light green	Dark Green
1N Hydrochloric acid	Light brown	Light green	Dark Green
Iodine	Greenish brown	Dark green	Dark bluish
Potassium hydroxide	Light Brown	Green	Light bluish
Ammonia	Light Brown	Green	Light bluish
1N Sodium hydroxide	Brown	Dark green	Dark bluish

TABLE 5: FLUORESCENCE ANALYSIS FOR SAMPLE 2

Solvent added	Colour observed under		
	Day light	Short UV wavelength (256 nm)	Long UV wavelength (365 nm)
1N Sulphuric acid	Light brown	Light green	Green
1N Nitric acid	Light brown	Light green	Green
1N Hydrochloric acid	Light brown	Light green	Green
Iodine	Greenish brown	Dark green	Dark bluish
Potassium hydroxide	Light Brown	Green	Light bluish
Ammonia	Light Brown	Green	Dark green
1N Sodium hydroxide	Brown	Dark green	Dark bluish

TABLE 6: HEAVY METALS TEST [14]

	Test	Observation	Inference
For Cadmium	NH_4OH added in the sample solution	White ppt. of cadmium hydroxide soluble in excess NH_4OH	Presence of cadmium
	Potassium ferrocyanide added	White ppt. of cadmium ferrocyanide.	Presence of cadmium
For Bismuth	H_2S gas added in the sample solution	Dark brown ppt. soluble in hot dil. HNO_3 but insoluble in NH_4S	Presence of bismuth
	NH_4OH	White ppt. insoluble in excess NH_4OH dissolved in dil. HCl	Presence of bismuth
For Lead	Dil. HCl added in sample solution	White ppt. of $CaCl_2$ soluble in boiled water and conc. HCl	Presence of lead
	KI is added in sample solution	Yellow ppt. soluble in boiling water	Presence of lead

TABLE 7: HEAVY METAL ANALYSIS

	Test	Observation	Result
Test for cadmium	NH_4OH added in the sample solution.	White ppt. is absent	Absence of cadmium
	Potassium ferrocyanide added	White ppt. is absent	Absence of cadmium
Test for bismuth	H_2S gas added in the sample solution	Dark brown ppt. is absent	Absence of bismuth
	NH_4OH	White ppt. is absent	Absence of bismuth
Test for lead	Dil. HCl added in sample solution	White ppt. of $CaCl_2$ is absent	Absence of lead
	KI is added in sample solution	Yellow ppt. is absent	Absence of lead

TABLE 8: QUALITATIVE PHYTOCHEMICAL SCREENING

	Test	Sample 1	Sample 2
Test of Tannin	5% $FeCl_3$ solution	Positive	Positive
	Lead acetate solution	Positive	Positive
	Bromine water	Positive	Positive
	Dilute iodine solution	Positive	Positive
Test for Alkaloids	Dragendroff's test	Positive	Positive
	Wagner's test	Positive	Positive
	Mayer' test	Positive	Positive

Detection of Alkaloids: 50 mg of solvent free extract was hydrolysed with dil. HCl and filtered. The filtrates were tested carefully with various alkaloid test reagents as follows.

Dragendroff's Test: To a few ml of filtrates, 1 to 2 ml of Dragendroff's reagent was added. A prominent yellow precipitate indicates the test is positive.

Wagner's Test: To a few ml of filtrates, few drops of Wagner's reagent were added by the side of the test tube. A reddish-brown precipitate confirms the test as positive.

Mayer's Test: To a few ml of filtrates, few drops of Mayer's reagent were added by the side of the test tube. A white or creamy precipitate if obtained indicates the presence of alkaloids.

RESULTS AND DISCUSSION: The churna was procured and was evaluated for its organoleptic, physical, physicochemical and fluorescence analysis. All the results obtained have been tabulated.

CONCLUSION: From the present investigation various standardization parameters such as physicochemical standards like total ash, acid insoluble ash, water and alcohol soluble extractive values, loss on drying, phytochemical analysis, flow properties and safety evaluation were carried out, it can be concluded that the formulation of Gandharva haritaki churna contains all good characters of an ideal churna and it was found to be harmless, more effective, and economic. The comparison between the one marketed sample and lab made churna have been done on the basis of the above mentioned parameters which shows satisfactory results, but the efficacy of the products can only be judged by doing the pharmacology of which is suggested as future scope of R & D.

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