



Research Article

PREPARATION AND EVALUATION OF PANCHA HARITHAKADI CHURNA FOR DIGESTIVE ACTIVITY

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ABSTRACT

Pancha Harithakadi Churna (PHC) is a traditional polyherbal formulation meant for digestion which consists of household ingredients having digestive property. PHC is mainly used for Constipation and Bloating. Churna's will play an important role in gastro intestinal problems. Churna's are having greater bioavailability because of smaller particle size. It consists of fine powder (sieve 100 size) of Ginger rhizomes, Fennel fruits, Myrobalan fruits, Senna leaflets and Pink Rock salt in equal proportions (1:1:1:1:1) are mixed well. PHC is formulated by standard procedures and evaluated by physical and analytical methods. Physicochemical standards and heavy metals are found to be within the limits. PHC is found to be free from microbes. The PHC showed pronounced amylolytic activity and moderate lipolytic and proteolytic activity when compared with standard showing its efficacy for treating indigestion. In future we will carry out other digestive enzymes *in vitro* studies and also to carry out *in vivo* digestive studies.

Key Words: PHC, Digestion, Physical evaluation, Ayurvedic formulation

INTRODUCTION

Churna is defined as a fine powder of drug or drugs in Ayurvedic system. Drugs are cleaned properly, dried thoroughly, pulverised and then sieved. It is a free flowing formulation and retains its potency for about one year. Triphala churna, Trikatu churna, Lavan Bhaskar churna are some of the examples. It is prescribed for treating diabetes, indigestion, ulcers, infections etc. Indigestion is a common problem but the usage of antacids may cause Alzheimer's disease upon long term usage. So, we prepared a churna with commonly used household ingredients^{1, 2}. The present study deals with the digestive activity of PHC. The ingredients of PHC are Ginger rhizomes (*Zingiber officinale*), Fennel fruits (*Foeniculum vulgare*), Myrobalan fruits (*Terminalia chebula*), Senna leaflets (*Cassia angustifolia*) and Pink Rock salt. Ginger contains Zingiberene³, Fennel contains Anethole³, Myrobalan contains Chebulic acid³, Senna contains Sennosides³ and Rock salt is also good for digestion. As per the literature we have formulated and evaluated for digestive activity namely Amylolytic, Lipolytic and Proteolytic activity.

MATERIALS & METHODS

Preparation of Churna

The raw materials such as Ginger rhizomes (1part), Fennel fruits (1part), Myrobalan fruits (1part), Senna leaflets (1part) and Pink Rock salt (1part) were used for the preparation of PHC. The raw materials of PHC were purchased from the market and authenticated by the Botany Department of Hindu College, Guntur, Market Centre, Andhra Pradesh – 522002, India based on the microscopical characters of powdered drugs. All the

ingredients were powdered separately, passed through sieve number 100 and mixed together in specified proportions. The churna was packed in an air tight glass container.

PHYSICO-CHEMICAL EVALUATION PARAMETERS^{4, 5}

Determination of Total Ash

About 2g of powdered drug was accurately weighed in a silica crucible, which was previously ignited and weighed. The powdered drug was spread as a fine layer on the bottom of the crucible. The crucible was incinerated at a temperature not exceeding 450°C until free from carbon. The crucible was cooled and weighed for constant weight.

Determination of Water-Soluble Ash

The ash obtained in the determination of total ash was boiled for 5 minutes with 25mL of water. The insoluble matter was collected on an ash less filter paper and washed with hot water. The insoluble ash was transferred into a tared silica crucible and ignited for 15 minutes at a temperature not exceeding 450°C. The weight of the insoluble matter was subtracted from the weight of total ash. The difference in weight was considered as the water-soluble ash.

Determination of Acid Insoluble Ash

The ash obtained as described in the determination of total ash was boiled with 25mL of hydrochloric acid for 5 minutes. The insoluble ash was collected on an ash less filter paper and washed with hot water. The insoluble ash was transferred into pre-weighed silica crucible and ignited for 15 minutes at a temperature not exceeding 450°C. The crucible was cooled, and the weight was considered as acid insoluble ash.

Loss on Drying

About 5g of the powdered drug was accurately weighed in a tared dish and dried in an oven at 100-105°C. It was cooled in desiccators and weighed again. The loss on drying was calculated with reference to the amount of dried powder taken.

Water Soluble Extractive

About 5g of air-dried coarsely powdered drug was macerated with 100mL of chloroform water (99.5mL of water + 0.5mL of chloroform) in a closed flask for 24 hours, shaking frequently during the last 6 hours and allowed to stand for 18 hours. It was then filtered rapidly, 25mL of the filtrate was evaporated to dryness in a tared flat-bottomed shallow dish dried at 105°C and weighed.

Ethanol Soluble Extractive

About 5g of air-dried coarsely powdered drug was macerated with 100mL of 90% ethanol in a closed flask for 24 hours, shaking frequently during the first 6 hours and allow standing for 18 hours. It was then filtered rapidly, 25mL of the filtrate was evaporated to dryness in a tared flat-bottomed shallow dish dried at 105°C and weighed.

Crude Fibre Content

2g of the Churna was 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.

DETERMINATION OF MICROBES^{6, 7}

1g of churna was dissolved in lactose broth and volume adjusted to 100mL with the same medium. About 10mL of sample was transferred into 100mL of Macconkey broth and incubated for 18-24 hours at 43-45°C. A subculture was prepared on a plate with Macconkey agar and incubated at 43-45°C for 18-24 hours. The growth of red colonies of gram negative rods appearing as reddish zones indicates the presence of *E. coli*, if not indicates the absence of *E. coli*.

DETERMINATION OF HEAVY METAL CONTENT⁸

The churna was subjected to heavy metal analysis by atomic absorption spectrophotometer (AAS). 0.3g of powdered sample preparation was placed in digestion tubes stirred with 5mL nitric acid and left overnight for complete digestion. Next day add 5mL nitric acid to each tube vessel closed with lid and heated in 150°C at oven for 2hrs till the solution became transparent. Cooled samples were transferred into 10mL volumetric flask. Added 4mL water to vessel, covered with lid. Diluted to volume with water and used for the estimation of heavy metal concentration.

The digested samples were then analyzed for Cd, Pb, Hg, As using a graphite furnace atomic absorption spectrophotometer (AAS; GBC 932+, AUSTRALIA). A single beam hollow cathode lamp (GBC) was used for estimation of Cd and Pb. Concentration of Hg was determined through AAS using cold vapor technique whereas air acetylene flame was used for determination of Arsenic (As) concentration.

All necessary precautions were taken to avoid any possible contamination of the sample as per the AOAC guidelines. The metal quantification was based on calibration curves, which were determined through a series of concentrations prepared using the chemical standard with 1mg/mL concentration. The concentration of the respective metals in sample was expressed as mg of metal/kg (ppm).

DETERMINATION OF DIGESTIVE ACTIVITY

Preparation of Extract

About 100mg of accurately weighed quantity of churna was extracted with 20% aqueous glycerol and phosphate buffer (pH 7.8) in 1:4 ratio and filtered. The filtrate was used as an enzyme source⁹. The standard sample was prepared like the test sample.

Amylolytic activity

Extract (1mL) of churna and standard were incubated separately for 15minutes at 27°C and added to 1mL of the substrate (soluble starch 1% in phosphate buffer). The enzyme reaction was interrupted by the addition of 2mL of DNS reagent and heated for 5minutes. The absorbance was measured at 520nm^{10, 11}.

Lipolytic Activity

Preparation of Substrate Solution

2mL of castor oil was, neutralized to pH 7 and stirred well with the 25mL of water in the presence of 100mg of bile salt (sodium taurocholate) till an emulsion was formed^{10, 11}.

Procedure

Take 20mL substrate and added 5mL phosphate buffer at pH 7. The contents were stirred slowly in magnetic stirrer and the temperature was maintained at 35°C. The electrodes of the pH meter were dipped in reaction mixture and the pH was adjusted to 7. The enzymes extract (0.5mL) was added immediately and pH recorded. The timer was set such that at zero time the pH was observed as 7. Then pH dropped by 0.2 units with addition of N/10 NaOH was noted. The pH was brought to initial value and was continued for 30 to 60 minutes. The volume of alkali consumed at each time was noted.

$$\text{Lipolytic activity} = \frac{\text{Volume of alkali} \times \text{Strength of alkali}}{\text{Weight of sample} \times \text{Time in minutes}}$$

Proteolytic activity

Preparation of Substrate Solution

200mL of boiled milk was treated with acetic acid till casein precipitates out. The precipitate was then removed, dried and powdered. One gram of prepared casein was diluted to 100mL using distilled water^{10, 11}.

Procedure

Take 1mL of substrate solution added to 1mL of 0.1M phosphate buffer (pH 7.6) and 1mL calcium chloride. To this 1mL crude enzyme extract was added and digestion stopped after 1hour of incubation with 3mL of 5% Trichloroacetic acid solution. After 10minutes precipitate was removed by centrifugation and one portion of supernatant was mixed with 5mL Lowry's reagent. The mixture was then stained with dilute Folin-Ciocalteu reagent (1:2) and optical density measured at a wavelength of 650nm. The proteolytic activity was then calculated from standard curve in milligrams of tyrosine. Protein estimated by standard method and results were given in milligrams of liberated tyrosine per milligrams of dissolved protein per hour at 37°C as specific activity.

RESULTS

The Amylolytic activity of a churna was found to be 0.38mg/mL at 1mg/mL and that of standard amylase was 0.31mg/mL. The values are represented in fig 1.

The Lipolytic activity was found to be 0.44mg/mL and the standard lipase was 0.58mg/mL. The values are represented in fig 2.

The Proteolytic activity was found to be 0.69mg/mL and that of standard protease was found to be 0.78mg/mL. The values are represented in fig 3.

Table 1. Physicochemical analysis of PHC

S.No.	Parameters	Observation
1.	Total Ash (% w/w)	16.7
2.	Acid Insoluble Ash (% w/w)	2.7
3.	Water Soluble Ash (% w/w)	14
4.	Crude Fibre (g)	8.55
5.	Water Soluble Extractive (% w/w)	0.96
6.	Alcohol Soluble Extractive (% w/w)	2.85
7.	Loss on Drying	11.24

Table 2. Heavy metal analysis of PHC

Heavy Metals	Pancha Harithakadi Churna (mg/kg)	Permissible limits (ADI/FDA values) (mg of metal per kg(ppm))
Lead (Pb)	0.70	10 mg/Kg
Arsenic (As)	0.35	0.5 ppm
Cadmium (Cd)	<0.1/ND	0.3 mg/Kg
Mercury (Hg)	<0.05/ND	1.0 ppm

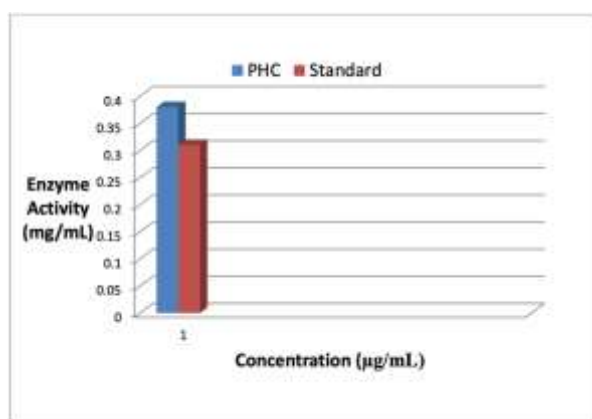


Fig 1: Determination of Amylolytic activity

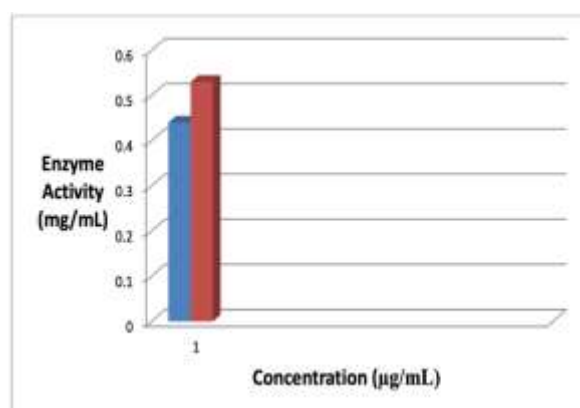


Fig 2: Determination of Lipolytic activity

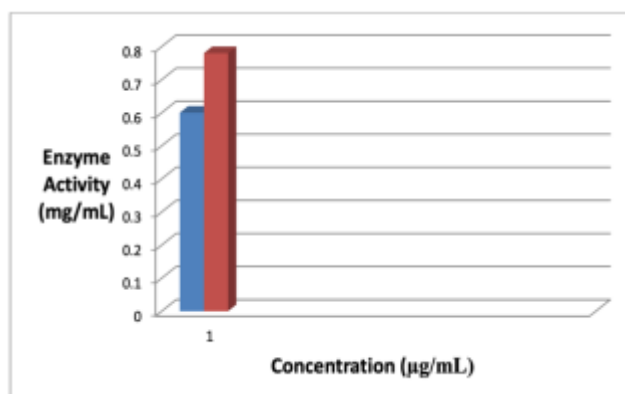


Fig 3: Determination of Proteolytic activity

DISCUSSION

PHC was subjected to standardisation by means of Physicochemical, Microbial and Heavy metal analysis. The Physicochemical standard values are within the standard limits. Heavy metals like Pb, As, Cd and Hg were estimated using AAS and the values are within the permissible limits. The churna was tested for *E. coli* and was found to be absent, hence the formulated churna complied with the WHO requirements.

The Digestive activity was evaluated by evaluating Amylolytic, Lipolytic and Proteolytic activity in comparison with the standard. The Amylolytic activity of a churna was found to be 0.38mg/mL at 1mg/mL and that of standard was 0.31mg/mL, Lipolytic activity was found to be 0.44mg/mL and the standard was 0.58mg/mL then Proteolytic activity was found to be 0.69mg/mL and that of standard was found to be 0.78mg/mL. In the present study the Amylolytic activity of formulated churna was found to be 7% greater than that of standard.

CONCLUSION

The Physicochemical parameters and Heavy metal analysis confirms the formulated churna complies with the WHO standards. The invitro digestive activity of PHC reveals the property of digesting carbohydrates, proteins and fats. Further investigation and isolation of compounds is necessary to establish the exact chemical constituents responsible for digestive activity.

REFERENCES

1. Natkarni AK, Indian Materia Medica, Popular Prakashan, 1976; 1, 800-806.
2. Cotton CM, Ethano botany principles and applications, Wiley and Sons Ltd., Chi Chestei, UK., 1996; 350.
3. Sukumar E, Phytochemistry and pharmacology of some famous Indian Medicinal plants, Vivekananda Kendra, Patrika, 1987; 04.
4. Jain sanjay, Koka Swetha, Gupta Asim, Barik Rakesh, Malavia Neelesh, Standardisation of Chopchiniyadi churna: An Ayurvedic formulation, Journal of Pharmacognosy, 2010; 2(50), 61.
5. Pattnayak P, Hardel DK, Mahapatra P, Standardisation of Vaisvanara Churna: A Polyherbal formulation, Journal of pharmacognosy, 2010; 2(5), 52.
6. Meena AK, Mangal AK, Rao MM, Panda P, Simha GV, Shakya SK et al., Evaluation of Standardisation parameters for Sitopaladi Churna an Ayurvedic Formulation, Asian Journal of research in Chemistry, 2011; 4(12):1867-1871.
7. WHO Quality control methods for medicinal plant materials, AITBS Publishers, Delhi 2002b, 65-67.
8. Indian Pharmacopoeia, Ministry of Health and Family Welfare, New Delhi, Government of India; 1996, 2, A 138-143.
9. Harold Varley, Practical clinical biochemistry, CBS Publishers 1988; 4, 245.
10. Chamundeeswari D, Kanimozhi P, Vasanth kumar M, Formulation and evaluation of churna for digestive property, Sri Ramachandra journal of medicine, 2017; 39-43.
11. Paul A, Wangano J, Patel G, Pharmacological invitro investigation for digestive property and invitro anti-ulcer activity of pep-up syrup, International journal of green pharmacy, April-June 2014; 119-124.

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