



Trade Science Inc.

Natural Products

An Indian Journal

Full Paper

NPAIJ, 3(2), 2007 [107-110]

Quantification Of Tannic Acid In Ayurvedic Formulation Bhuvneshavara Vati: Spectrophotometric Approach For Routine Quality Control

V.Jain, Swarnlata Saraf, S.Saraf*

Institute of Pharmacy, Pt. Ravishankar Shukla University, Raipur(C.G.), 492 010, (INDIA)

E-Mail: vj_rsofiop@rediffmail.com

Received: 29th May, 2007 ; Accepted: 3rd June, 2007

ABSTRACT

A widely used Ayurvedic preparation, 'Bhuvneshavara Vati' has been prepared as per Ayurvedic formulary of India was estimated spectrophotometrically for its tannic acid content. Three-laboratory batch of Bhuvneshavara Vati were estimated for their tannic acid contents against standard tannic acid solution on double beam UV-Visible Spectrophotometer at λ_{max} 276nm. The concentration of tannic acid present in raw material was found to be $6.1\% \pm 0.27w/w$ in amla, $8.7\% \pm 0.31w/w$ in bahera, $14.05\% \pm 0.29w/w$ in harda, $4.8\% \pm 0.94$ in bael and 0.67 ± 0.49 in ajowan and in three identical laboratory batch of Bhuvneshavara Vati name BV-I, BV-II, BV-III, was $4.90\% \pm 0.42$, $4.79\% \pm 0.86$, $4.85\% \pm 0.75w/w$ respectively with mean value $4.85\% \pm 0.53 w/w$. The tannic acid content of all the three batches is found to be in close proximities with each other and recovery studies are indicative of reproducibility of method. Hence the present method is simple, sensitive, precise and accurate and can be adopted for routine quality control of Bhuvneshavara Vati. © 2007 Trade Science Inc. - INDIA

KEYWORDS

Tannic acid;
Bhuvneshavara vati;
UV;
Fingerprinting;
Ayurvedic formulation;
Quality control parameter.

INTRODUCTION

The most of the Ayurvedic formulation are lacking in their defined quality control parameters and method of its evaluation^[1]. The World Health Organization(WHO) in its resolution WHA 31.33(1978), WHA 40.33(1987), WHA 42.43(1989)

has emphasized the need to ensure the quality of medicinal plant products by using modern controlled technique and applying suitable standards^[2]. The present paper is an effort to develop the quality control parameter of Bhuvneshavara Vati by spectrophotometric determination using tannic acid as an internal standard.

Full Paper

Bhuvneshavara Vati among the most common formulas used for diarrhea (Atisara) in Ayurvedic medicine. Comprised of the fruits of five medicinal important plants, Indian gooseberry (Amalaki, *Emblica officinalis*), Belleric myrobalan (Vibhitaka, *Terminalia belerica*), Chebulic myrobalan (Haritaki, *Terminalia chebula*), Yamani (Ajowan, *Trichyspermum ammi*), Bilvapesika (Bael, *Aegle marmelos*) and two mineral ingredients saindhava lavan (rock salt), grahadhoom (Soot)^[3].

In this connection an attempt has been made to develop the method of estimation of tannic acid which is chemically hydrolysable tannin corresponding to a complexity of pentadigalloyl glucose^[4] C₇₆ H₅₂ O₄₆ an important content in Bhuvneshavara Vati. Present study is based on UV spectrophotometric analysis, which is a simple, precise, and accurate method that can be considered as quality control method for routine analysis.

EXPERIMENTAL

Plants

All the crude drugs were purchased from local market Raipur (C.G.), India and identified on the basis of morphological and microscopical characters and compared with standard Pharmacopoeial Monograph^[5-8].

Chemicals

All the chemicals and solvents were used of A.R. Grade.

Preparation of Bhuvneshavara Vati

Bhuvneshavara Vati, three batch name BV-I, BV-II, BV-III, were prepared in laboratory using method described in Ayurvedic Formulary^[10]. The individual fruit of amla, bahera, harda, ajowan and bael was also powdered. These three batches of Bhuvneshavara Vati and powdered amla, bahera, harda, ajowan and bael were estimated for their tannic acid contents against standard tannic acid solution on UV-Visible Spectrophotometer (Shimadzu, UV-1700, Pharmaspec). As mineral ingredients saindhava lavan (rock salt) along with grahadhoom (Soot) does not contain tannic acid is not included in present study.

Preparation of tannic Acid extract of Bhuvneshavara Vati

The tannic acid extract for each batch of Bhuvneshavara Vati and separately powdered amla, bahera, harda, ajowan and bael were prepared as per the method described by Jain et al.⁹.

Preparation of standard solution of tannic acid

As tannic acid show good solubility in 0.1N Hydrochloric acid, an accurately weighed tannic acid (100mg), from Himedia, A.R. Grade, was dissolved in 0.1N hydrochloric acid and volume was made up to 100ml with 0.1 N hydrochloric acid in volumetric flask. 2ml of this solution was diluted with 0.1 N hydrochloric acid up to 100 ml in volumetric flask to give 20 µg/ml tannic acid stock solution.

Calibration Curve of tannic acid

A series of calibrated 10 ml volumetric flask were taken and appropriate aliquots of the working standard solution of tannic acid were withdrawn and diluted up to 10ml with 0.1 N hydrochloric acid. The absorbance was measured at absorption maxima 276nm, against the reagent blank prepared in similar manner without the tannic acid. The absorption maxima and Beer's law limit were recorded and data that prove the linearity and obey Beer's law limit were noted.

The linear correlation between these concentrations (X-axis) and absorbance (Y-axis) were graphically presented and the slope (b), intercept (a), and correlation coefficient (r²) were calculated out for linear equation (Y = bx + a) by regression analysis using the method of the least square, TABLE 1 and figure 1.

Determination of limit of quantitation and limit of detection

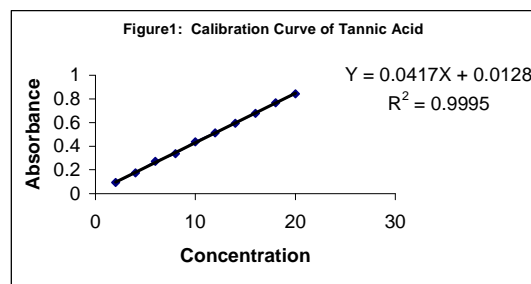


Figure 1: Calibration curve of Tannic Acid

TABLE 1 : Optical characteristics, statistical regression data and validation parameter of Tannic Acid

S.No.	Parameter	Value
1	Absorption Maxima	276 nm
2	Beer's Law limit	2-20µg/ml
3	Regression equation (y=bx+a)	y=0.0417x+0.0128
4	Intercept (a)	0.0128
5	Slope (b)	0.041682
6	Correlation coefficients (r ²)	r ² =0.9995
7	Precision (n=6, % RSD)	0.251
8	Accuracy (%)	99.30
9	LOQ	0.362µg/mL
10	LOD	0.129µg/mL

TABLE 2 : Estimation of tannic acid content in Bhuvneshavara Vati

S.no.	Name	Tannic acid content %w/w	Confidence level (95%)
1	Amla	6.1% ± 0.27	±0.216
2	Harda	14.05%± 0.31	±0.248
3	Bahera	8.7% ± 0.29	±0.232
4	Bael	4.8%±0.94	±0.796
5	ajowan	0.67%±0.49	±0.412
6	Bhuvneshavara BV-I	4.90%±0.42	±0.402
7	Bhuvneshavara BV-II	4.79%±0.86	±0.722
8	Vati BV-III	4.85%±0.75	±0.644

Mean±SD of six determinations, BV-I: Bhuvneshavara Vati Batch I, BV- II: Bhuvneshavara Vati Batch II, BV-III: Bhuvneshavara Vati Batch III

TABLE 3 : Compilation of data of recovery study

S.No	Amount of Tannic Acid (µg/ml)			RSD%	SE	Recovery%
	In sample	Added	Estimated			
1	100	50	149.23±0.39	0.261	0.159	99.48±0.26
2	100	100	198.25±0.48	0.242	0.196	99.13±0.24
Mean				0.251	0.77	99.30

Mean ± SD of six determinations, RSD=Relative standard deviation, SE=Standard error

The limit of detection (LOD) is the lowest amount of analyte in a sample which can be detected but not necessarily quantitated as an exact value. The limit of quantitation (LOQ) is the lowest amount of analyte which can be quantitatively determined with suitable precision. The LOD and LOQ of the developed method were determined by injecting progressively low concentration of the standard solu-

tion and the lowest concentrations assayed.

Estimation of tannic acid:

The appropriate aliquots from tannic acid extract of each batch of Bhuvneshavara Vati & separately amla, bahera, harda, ajowan and bael were withdrawn in 10ml volumetric flask separately absorbance for aliquots of each was noted at 276nm. The corresponding concentration of tannic acid against respective absorbance value was determined using the tannic acid calibration curve. The statistical analysis for checking uniformity in batches is also performed (TABLE 2)

Precision and accuracy

The method was validated for precision and accuracy, by performing the recovery studies at two levels by adding known amount of tannic acid extract of Bhuvneshavara Vati, of which the tannic acid content have been estimated previously. The data were obtained and recovery was calculated (TABLE 3).

RESULTS AND DISCUSSION

Tannic acid was found obey Beer Lambert's law in concentration range 2-20µg/ml at λ_{max} 276 nm. The correlation coefficient (r²) was calculated where the r² value 0.9995 indicates the good linearity between the concentration and absorbance. Under the developed UV conditions, the limit of quantitation was determined to be 0.362µg/mL with triplicate of the sample. Also, the limit of detection was found to be 0.129µg/mL.

The estimation of tannic acid content of Bhuvneshavara Vati (three identical laboratory batch) and powdered amla, bahera, harda, ajowan and bael was carried out separately. The concentration of tannic acid present in raw material was found to be 6.1%±0.27w/w in amla, 8.7%±0.31w/w in bahera, 14.05%±0.29w/w in harda, 4.8%±0.94 in bael and 0.67±0.49 in ajowan respectively and in three identical laboratory batch of Bhuvneshavara Vati name BV-I, BV-II, BV-III, was 4.90%±0.42, 4.79%±0.86, 4.85%±0.75w/w (TABLE-2) respectively with mean value 4.85%±0.53 w/w.

Full Paper

In order to obtain Precision and Accuracy the Recovery study was performed at two levels by adding known amount of tannic acid with preanalysed sample of tannic acid in Bhuvneshavara Vati. The experiment was repeated six time at both level (TABLE 3) and result shows $99.48\% \pm 0.26$ and $99.13\% \pm 0.24$ recovery of tannic acid at both the level with mean value $99.30\% \pm 0.25$ which prove reproducibility of the result. This shows significant Precision of methods with 95% confidence level. The %Relative Standard Deviation(%RSD) value was found to be 0.261 and 0.242 with mean 0.251 at both the level while the Standard Error was 0.159 and 0.196 with mean 0.177 respectively. From the data's it is obvious that the present method of Spectrophotometric determination of tannic acid is simple, precise, accurate and suitable for routine analysis of tannic acid in Bhuvneshavara Vati.

As Bhuvneshavara Vati is a good source of tannic acid, these findings can be taken as one of the parameter, along with other parameters, for Quality control of Bhuvneshavara Vati.

ACKNOWLEDGEMENT

The authors are highly grateful to Director, Institute of Pharmacy, Pt. R.S. University, Raipur and AICTE for providing financial assistance under Research Promotion Scheme (AICTE-RPS).

REFERENCES

- [1] V.Jain, S.Saraf, S.Saraf; Asian journal of chemistry, **19(7)**, (2007).
- [2] World Health Organization, Quality Control Methods For Medicinal Plants Materials, Geneva, 1-15 (1998).
- [3] The Ayurvedic Formulary of India, Part II, 1st english edition, Govt.of India, Ministry of Health and Family Planning, Dept. of Indian system of medicine and Homeopathic, Delhi, 178 (2000).
- [4] N.S.Khan, S.M.Hadi; Mutagenesis, **13(3)**, 271-4 (1998).
- [5] The Ayurvedic Pharmacopoeia of India, Part I, Ministry Of Health And Family Welfare, Government Of India, Department of Health, New Delhi, **1(1)**, 4,5,26,47,48 (1986).
- [6] Indian Herbal Pharmacopoeia, Regional Research Laboratory Jammu, Indian drug Manufacturing Association Mumbai, **2**, 50-57 (1999).
- [7] Quality Standards of Indian Medicinal Plants, Indian Council of Medicinal Research, New Delhi, **1**, 198-212 (2003).
- [8] P.Mukherjee; Pharmacological Screening of Herbal Drug, 'Quality Control of Herbal Drug: An Approach to Evaluation of Botanicals', Eastern Publishers (Business Horizontal Ltd.), New Delhi, 539-541 (2002).
- [9] V.Jain, S.Saraf, S.Saraf; Asian Journal of Chemistry, **19(2)**, 1406-1410 (2007).