IJPSR (2018), Volume 9, Issue 10 (Research Article)

INTERNATIONAL JOURNAL OF PHARMACEUTICAL SCIENCES AND RESEARCH

Received on 22 January, 2018; received in revised form, 26 April, 2018; accepted, 13 May, 2018; published 01 October, 2018

EFFECT OF PROPORTION COMPOSITION VARIATION ON PHYSICOCHEMICAL PARAMETERS OF TRIPHALA

Avinash Kondalkar [1], Sapna Avinash Kondalkar [*2], Vijay Kumar [2], Anupam K. Mangal [3] and V. Subhose [2]

Sun Institute of Pharmaceutical Education and Research [1], Lahar - 477445, Madhya Pradesh, India.
Regional Ayurveda Research Institute for Drug Development [2], Gwalior - 474009, Madhya Pradesh, India.
Central Council for Research in Ayurvedic Sciences [3], New Delhi - 110058, Delhi, India.

Keywords:

Triphala Churna, *Emblica officinalis*, *Terminalia chebula*, *Terminalia bellerica*

Correspondence to Author:
Sapna Avinash Kondalkar

Research Officer Pharmacognosy, Regional Ayurveda Research Institute for Drug Development (CCRAS), Aamkho, Gwalior - 474009, Madhya Pradesh, India.

E-mail: sapna.soni@rediffmail.com

ABSTRACT: Background: Triphala is exclusively used in more than 200 drug formulations in Indian system of Medicine. The formulation process of polyherbal drugs changes the physiochemical properties of the whole drug which need to be studied and understood. **Objective:** The present work aims to perform the organoleptic features (color, odor, taste), and physicochemical parameters such as pH, loss on drying, ash values *viz.*, total ash, acid insoluble ash, water-soluble ash, extractive values *viz.*, alcohol soluble extractive and water-soluble extractive tests and were determined under variable ratio concept. **Materials and Methods:** The standard methods as per traditional text were followed and the physicochemical changes were also investigated. Study of individual fruit as well "under variable ratio" concept (Amla: Bahera: Harad (1:1:1), (2:1:1), (1:2:1), and (1:1:2)) of three fruits were performed. Statistical analysis was performed at significant level $p \leq 0.05$. **Results:** The observed range of various tests were as: pH (2.5-5.9), loss on drying (4.48-13.71), total ash (2.92-10.80), acid insoluble ash (0.53-4.92), extractive values *viz.*, alcohol soluble extractive (39.02-73.03) and water-soluble extractive (12.60-54.42). The physicochemical analysis results of four proportions were statistically significant at $p \leq 0.05$. **Conclusion:** Current study has revealed the variation of physicochemical parameters with ratio variation. Hence this study would be very much helpful for the identification and standardization of Triphala churna to differentiate from other powdered sources. Most importantly, ratio variation of Triphala had shown physicochemical parameters variation which is big tool for quality control in future.

INTRODUCTION: Even today in Western medicine, and despite progress in synthetic chemistry, plants are the backbone of primary health care and approximately 80% population still relies upon plants. Herbal medicines trends are growing quickly and became a major part of the economy (60 million USD) [1].

QUICK RESPONSE CODE

DOI: 10.13040/IJPSR.0975-8232.9(10).4280-85

Article can be accessed online on: www.ijpsr.com

DOI link: http://dx.doi.org/10.13040/IJPSR.0975-8232.9(10).4280-85

However, Ayurveda is based on two principles, use as single drug or use more than a drug. Several drugs are combined to achieve extra therapeutic effectiveness and known as polyherbal formulations [2, 3].

The concept of polyherbalism was mentioned historically in the Ayurvedic literature "Sarangdhar Samhita" in 1300 A.D. Due to synergism, polyherbal formulations offer some benefits which are not available with single drugs. Despite their benefits some drawbacks are still there, like the source, poor manufacturing process, manufacturers' irresponsibility, quality aspects, laws and regulations *etc*.

If preventive and corrective measures taken, Ayurvedic polyherbal formulations can exert the best effect to treat diseases and to maintain human health [2, 3].

Polyherbal drug, Triphala powder is an Ayurvedic formulation consisting of powders of three fruits, amalaki (*Emblica officinalis*), haritaki (*Terminalia chebula*), and bibhitaki (*Terminalia belerica*) in equal proportions [3, 4, 5]. The churnam is exclusively used in multiple drug formulations in Indian system of medicine. Triphala is exclusively used in more than 200 drug formulations in Indian system of Medicine [4, 5, 6, 7, 8]. Three plants, amalaki (*Emblica officinalis*), haritaki (*Terminalia chebula*), and bibhitaki (*Terminalia belerica*) distributed throughout the greater part of India, Burma and Srilanka [3, 4, 5, 6, 7]. The Triphala churna is found to be an effective astringent and used in diarrhoea, dysentery and extensively used as additive in systemic treatments of diabetes, spasm, scurvy, jaundice, dyspepsia, cough, asthma, diseases of the eye, diarrhoea *etc*. [3, 4, 5, 6, 7, 8, 9, 10, 11].

As all the ingredients possess specific characteristic, improper formulation may result variation in the therapeutic effect. According to WHO, phytochemical and physico-chemical characterization is necessary to determine its identity, quality and purity standard and to ensure reproducible quality products [10, 11, 12, 13]. Physical standards are constant for crude drugs, but vary due to the presence of adulterants or improper processing [12]. Hence, in the present study we have performed physico-chemical characterization of Triphala churna, where we have checked the effect of ratio variation on the physico-chemical properties of Triphala churna. To our knowledge, this is the first study to perform physico-chemical tests of Triphala churna under variable ratio concept.

MATERIALS AND METHODS:

Procurement and Preparation: Fruits were purchased from the local market of Gwalior, India, extraneous matter was removed and were cut into small pieces, dried and packed in air- tight polyethylene bags. All the dried fruits were pulverized separately, passed through sieve (80 mesh) for uniform and desired particle size, packed and labelled.

Different samples of variable ratios were prepared by mixing amla, bahera and hard fruit powders as shown in the **Table 1**. All the samples were packed in air- tight self-sealing polyethylene bags, labelled (I-VII) and keep in desiccator for further evaluation [11, 12, 13].

TABLE 1: DIFFERENT SAMPLES AND THEIR COMPOSITION

Sample Code	Composition
I	Amla fruit powder
II	Bahera fruit powder
III	Harad fruit powder
IV	Amla: Bahera: Harad (1:1:1)
V	Amla: Bahera: Harad (2:1:1)
VI	Amla: Bahera: Harad (1:2:1)
VII	Amla: Bahera: Harad (1:1:2)

Determination of Organoleptic Features: All prepared samples (I-VII) were evaluated for color, odor and taste as per the recent protocol [13, 14, 15].

Determination of Loss on Drying: 4 g of the sample was taken in a previously weighed 100 ml beaker and heat in an oven at 105 ºC for 5 h. Beaker was kept in a desiccator for cooling and weighed. Procedure was repeated till two constant observations [2, 13]. Percentage of loss in weight of the sample was calculated by using formula given below.

$$\text{Percentage of loss on drying at 105 ºC} = \frac{\text{Loss in weight of the sample}}{\text{Weight of the sample by taken}} \times 100$$

Determination of Total Ash: 4 g of the sample was taken accurately in a previously ignited and tarred silica dish. Material in dish was spread evenly and ignite in a muffle furnace at 600 ºC until it is white, indicating the absence of carbon. Dish was kept in a desiccator until cooling and weighed [2, 13]. Percentage of total ash of the sample was calculated by using formula given below.

$$\text{Percentage of total ash} = \frac{\text{Weight of ash}}{\text{Weight of sample taken}} \times 100$$

(Note: If carbon free ash cannot be obtained in this manner, cool the dish and moisten the residue with about 2 ml of water or saturated solution of ammonium nitrate).

Determination of Acid- insoluble Ash: To the dish containing the total ash, 45 ml (of 1: 5 hydrochloric acid in three portions of 15 ml each time) was added, boiled gently for 5 min and filtered. The insoluble matter was collected on an

ash less filter paper (Whatmann no. 41) and washed with distilled water until the residues were free from acid. The filter paper containing the insoluble matter was transferred to the original dish, dried and ignited to constant weight [2, 13]. After cooling the dish in a desiccator, the percentage of Acid-insoluble ash of material was calculated as:

$$\text{Percentage of Acid-insoluble ash} = \frac{\text{Weight of the Acid - insoluble residue}}{\text{Weight of the sample}} \times 100$$

Determination of Water- soluble Extractive: 4 g of the sample was taken in a glass stopper flask. 100 ml of distilled water was added, and shaken occasionally for 6 h, and then allowed to standing for 18 hours. Extract was filtered rapidly taking care not to lose any solvent. 25 ml of the filtrate was pipette out in a pre-weighed 100 ml beaker and evaporated to dryness on a water bath. Further, beaker was kept in a hot air oven at 105 ºC for 6 h, cooled in a desiccator and weighed. Experiment was repeated twice, and resulted in average value [2, 13]. The calculation was done by using formula given below.

$$\text{Percentage of water -soluble extractive} = \frac{\text{Weight of the extract} \times 100 \times 100}{25 \times \text{weight of the sample taken}}$$

Determination of Alcohol - soluble Extractive: Same procedure was repeated as that of determination of water - soluble extractive value. The change was made by replacing water with absolute alcohol (approximately 95%) [2, 13].

Determination of pH: Calibrated pH meter was used to determine the pH value. 10% solution of sample (I-VII) was introduced one by one and reading was noted. Test was repeated for three times, and the average reading reported as a result [2, 13].

Statistical Analysis: All the quantitative tests were performed in replicated. The results are presented as ± mean of replicates. All the data were analyzed using the Excel Window 8 version software. Statistical analysis was done by analysis of variance (ANOVA) followed by Tukey's test. $p \leq 0.05$ was considered to be statistically significant.

RESULTS AND DISCUSSION: Among the organoleptic features the observed color was between yellowish to yellowish brown **Table 2**. The observed odor was characteristic or/and astringent for maximum samples. Same thing was noticed for taste features **Table 2**. The observed organoleptic features were same as that of market samples study which was performed by Kadam *et al*., 2017 [16]. These sensory traits were the most influential traits studied to pronouncing as a "decision on preliminary step". These primarily studies are very important w.r.t. shelf life and proper preparation of any herbal drug and food supplement [13, 14, 15]. Foul smelt and bad taste food/herb/supplements are never considered as good product for health. Most significantly, among raw materials, these organoleptic and sensory features can vary w.r.t. various conditions including way of drying, storage, geographical conditions *etc*. [14, 15, 16]

TABLE 2: ORGANOLEPTIC FEATURES OF SAMPLES (I-VII)

Sample Code	Color	Odor	Taste
I	Light yellowish brown	Sore, characteristic	Characteristic, sore, cooling-sweet after- taste
II	Pale brown	Faint characteristic, astringent	Characteristic, astringent
III	Yellowish brown	Faint characteristic, astringent	Characteristic, astringent
IV	Light yellowish brown	Astringent, characteristic	Sore, astringent, characteristic
V	Yellowish brown	Sore, characteristic, astringent	Characteristic, sore, astringent
VI	Light pale brown	Characteristic, astringent	Characteristic, astringent
VII	Pale brown	Characteristic, astringent	Characteristic, astringent

Physicochemical parameters of three ingredients as well as four formulations (of variable proportion) of Triphala were determined **Table 3**. Powder of the three fruits were analyzed as single drug along with four formulation prepared with Amla: Bahera: Harad in varying proportions [(1:1:1), (2:1:1), (1:2:1), and (1:1:2)]. The observed range of various physicochemical tests were as: pH (2.5-5.9), loss on drying (4.48-13.71), total ash (2.96-11.23), acid insoluble ash (0.53-4.92), extractive values *viz.*, alcohol soluble extractive (39.02-73.03)) and water - soluble extractive (12.60-54.42). Comparatively, the results of samples I-VI were matched with the Ayurvedic Pharmacopeial Standards, and were

within the limits. But, slight higher values were noticed for bahera fruit. On other hand, till date no study has been performed for variable proportions. Hence these are very new results for sample number V-VII. It was observed that if the proportion changes, the physicochemical parameters changes. Small changes were noticed for amla and harad, but the remarkable changes were noticed as the proportion of bahera changed **Table 3**. The physicochemical parameters of three fruits or first three samples of present study was very similar to study performed by Shivakumar *et al*., 2016 [17]. Similarly, the physicochemical parameters of Triphala churn (1:1:1) or VIth samples of present study was very similar to study performed by Sangolgi *et al*., 2017 [18].

The observed physic-chemical parameters of Triphala churna (1:1:1) were much close to the recent reports. Ashok Kumar has reported that the physic-chemical parameters of Triphla churna as: total ash (10.21 ± 0.42), acid insoluble ash (2.54 ± 0.06) and the extractive values *viz*. water soluble extractive (52.56 ± 2.04) and alcohol soluble extractive (11.20 ± 0.18) were recorded [9]. In present study, the value of water soluble extract was varied significantly. It might me because of change in geographical conditions. Kadam *et al*., performed a comparative study on marketed Triphala churna samples and they have noticed that the extractive value of samples were 1 - 3%, loss on drying (0.71 - 2.0%), and ash content (1 - 3%) respectively [16]. These results were significantly different than current study. As like that of Kadam *et al*., other research group of Bahuguna *et al*., 2014 has observed same results for marketed formulation [16, 19]. Yogesh *et al*., has reported the water and alcohol extractive values 44.72 and 36.37% respectively [20].

Water soluble extractive value was similar to current study, but huge difference was notice for alcohol soluble extractive values of Triphala churn. Parameters analyzed in current study for Triphala churna (1:1:1) were almost closed to the results published by Kaushika *et al*., 2015, except the alcohol soluble extract which was reported 8 - 11% [21]. The slight percentage variation among the physicochemical parameters and chemical constituents of plants belongs to geographical conditions. Recently, Yadav has reported the variation of tannin in triphala drugs and triphala churna with regional variations [22].

TABLE 3: PHYSICOCHEMICAL PARAMETERS FOR DIFFERENT SAMPLES (I-VII)

Sample	LOD (%)*	Total Ash (%)*	Acid-Insoluble Ash (%)*	Water-soluble Extractive (%)*	Alcohol-soluble Extractive (%)*	pH*
I	4.48	3.43	1.53	67.01	54.42	2.5
II	6.76	7.59	0.93	48.36	29.07	4.5
III	5.31	2.96	4.41	73.03	51.91	3.2
IV**	11.86	9.71	2.81	45.50	13.96	5.5
V	11.90	10.72	4.63	48.30	14.23	5.6
VI	13.71	11.23	3.63	39.02	12.60	5.4
VII	12.50	10.67	4.92	54.26	14.98	5.9

*Here all the results are represented as average vale, ** sample IV taken as standard and compared with sample V-VII at p≤0.05.

Statistical analysis of sample IV-VII were performed by considering sample IV (1:1:1) as standard. At the $p \leq 0.05$ level, the means of loss on drying, pH, total ash, acid insoluble ash, water soluble extractive value, and alcohol soluble extractive value were significantly different. The loss on drying percentage variation ($p \leq 0.05$) for sample V, VI, and VII were 0.34, 15.59, and 5.39 respectively **Fig. 1A**. The total ash content percentage variation ($p \leq 0.05$) for sample V, VI, and VII were 10.72, 11.23, and 10.67 respectively **Fig. 1B**. The acid insoluble-ash content percentage variation ($p \leq 0.05$) for sample V, VI, and VII were 64.76, 29.18, and 75.08 respectively **Fig. 1B**. The water soluble extractive value percentage variation ($p \leq 0.05$) for sample V, VI, and VII were 6.15, -14.24, and 19.25 respectively **Fig. 1C**. The alcohol soluble extractive value percentage variation ($p \leq 0.05$) for sample V, VI, and VII were 1.93, -9.74, and 7.30 respectively **Fig. 1C**.

The pH value percentage variation ($p \leq 0.05$) for sample V, VI, and VII were 1.81, -1.81, and 7.27 respectively **Fig. 1D**. In statistical study, the maximum variation was noticed for sample VI having 1:2:1 (Amla: Bahera: Harad), followed by

sample VII (1:1:2) and V (2:1:1). The current study is a strong clue for the good laboratory practice while preparing the polyherbal drugs. Current study is highlighting that once ratio is varied, all physiochemical parameters would be changed, which leads to variations in efficacy and therapeutic index of drug, especially of polyherbal drugs.

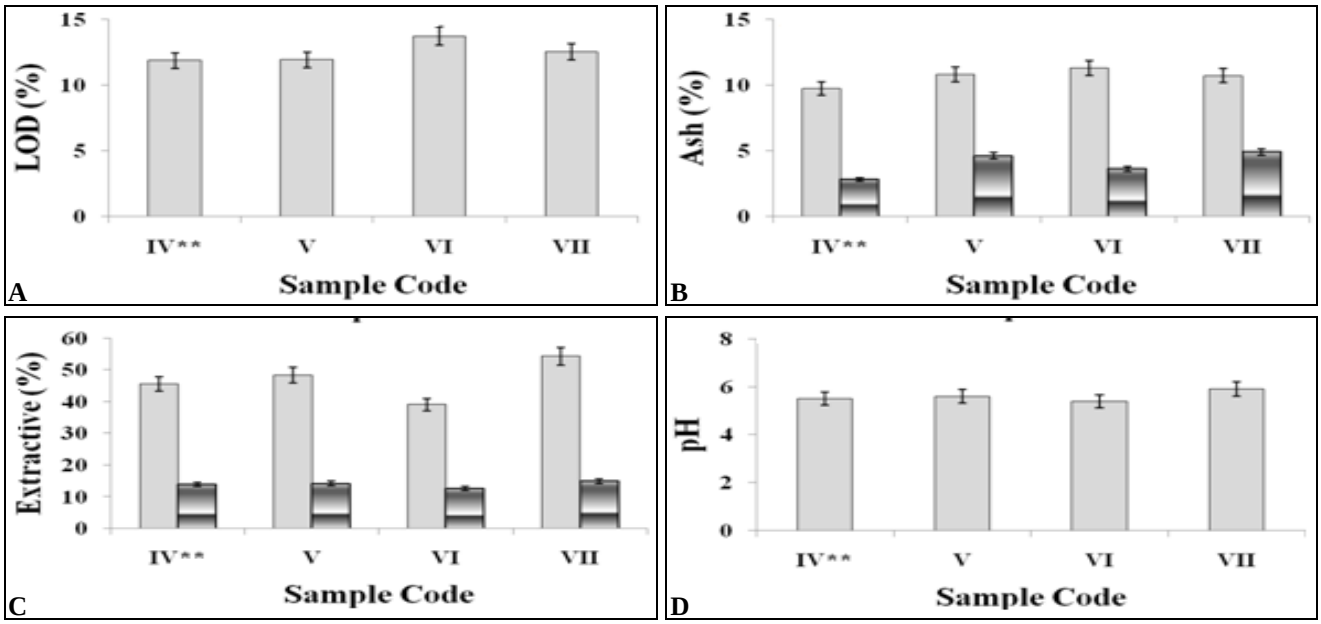

FIG. 1: 1A: LOD (%), 1B: ASH (%) (▭ TOTAL ASH; ▭ ACID INSOLUBLE ASH), 1C: EXTRACTIVE VALUES (%) (▭ TOTAL ASH; ▭ ACID INSOLUBLE ASH), AND 1D: PH VALUES, OF FOUR DIFFERENT PROPORTIONS SAMPLES OF TRIPHALA CHURN AT SIGNIFICANTLY LEVEL P≤0.05

CONCLUSION: As the therapeutic effect of a poly- herbal formulation is a synergistic effect of specific characteristics of individual ingredient, improper formulation may result variation in the therapeutic effect. Now a day, it is necessary to cop up with the regulatory requirements. This preliminary study could be helpful in analyzing the composition of Triphala Churna and to check the deviation from the formula. In present study, at significant level ($p \leq 0.05$), there was variation among all physiochemical parameters were noticed with ratio variation. This investigation is very much helpful for the identification of Triphala churna composition, which would be useful in the field of clinical, pharmacognosy, phytochemistry, botany and herbal industry for further research activities.

ACKNOWLEDGEMENT: We are grateful to Prof. Vd. K. S. Dhiman [(Director General Central Council for Research in Ayurvedic Sciences (CCRAS) is an autonomous body of the Ministry of Ayush (Ayurveda, Yoga and Naturopathy, Unani, Siddha and Homeopathy), Government of India)] for his huge encouragement. We are also thankful to CCRAS for funding facilities and RARIDD for the library, and laboratory facilities.

CONFLICT OF INTEREST: The authors declare that there are no conflicts of interest.

REFERENCES:

1. Newman DJ and Cragg GM: Natural Products as Sources of New Drugs from 1981 to 2014. J. Nat. Prod. 2016; 79; 629-661.
2. The Ayurvedic Pharmacopoeia of India (API), Part I New Delhi: Government of India, Ministry of Health and Family Welfare, Department of Indian System of Medicine and Homeopathy 2008; 5(1): 227-228.
3. Parasuraman S, Thing GS and Sokkalingam AD: Polyherbal formulation: concept of Ayurveda. Pharmacog. Review 2014; 8; 73- 80.
4. Pal NS, Nair AS, Nair AM and Murali M: Pharmacological and therapeutic effects of triphala – A literature review. J Pharmacog. Phytochem 2016; 6; 11-19.
5. Bali C, Kumavat RC, Mita K, Ramamurthi A and Sumit N: Triphala: A comprehensive ayurvedic review. Int. J. Res. Ayu. Pharma. 2013; 4: 612- 617.
6. Kumar NS, Nair AS, Nair AM and Murali M: Pharmacological and therapeutic effects of triphala – A literature review. J Pharmacog. Phytochem 2016; 5: 23-30.
7. Santoshkumar J, Manjunath S and Sakhare PM: A study of anti-hyperlipidemia, hypolipedimic and anti-antherogenic activity of fruit of *Emblica officinalis* (amla) in high fat fed Albino Rats. Int. J. Med. Res. Health Sci. 2013; 2: 70-77.
8. Abraham A, Mathew L and Samuel S: Pharmacognostic studies of the fruits of *Terminalia bellirica* (Gaertn). Roxb," J Pharmacog. Phytochem 2014; 3: 45- 52.
9. Ashok KD: Pharmacognostical investigations on Triphala churnam. Ancient Sci. Life 2007; 26: 40-44.

10. Anand KK, Singh B, Saxena AK, Chandan BK, Gupta VN and Bhardwaj V: 3,4,5-Trihydroxy benzoic acid (gallic acid), the hepatoprotective principle in the fruits of *Terminalia belerica* bioassay guided activity. Pharmacological Res. 1997; 36(4): 315-321.
11. Mathur R, Sharma A, Dixit VP and Varma M: Hypolipidaemic effect of fruit of *Emblica officinalis* in cholesterol. J. Ethnopharmacol. 996; 50(2): 61-68.
12. Lohar DR: Protocol for testing Ayurvedic, Siddha and Unani Medicines. Pharmacopoeial Laboratory for Indian Medicines, Department of AYUSH, Ministry of Health and Family Welfare, Govt. of India, Ghaziabad 2005, 49-50.
13. WHO: WHO Guidelines for Assessing Quality of Herbal Medicines with Reference to Contaminants and Residues. Switzerland, Geneva 2007.
14. Chauhan VS and Sharma A: Studies on organoleptic properties of food products from fresh egg and egg powder through principal component analysis. Nahrung/Food 2003; 2: 102-105.
15. Yi J, Zhou L, Bi J, Chen Q, Liu X and Wu X: Influence of pre-drying treatments on physicochemical and organoleptic properties of explosion puff dried jackfruit chips. J. Food. Sci. Technol 2016; 53: 1120-9.
16. Kadam DK, Ahire PD, Bhoye JV, Patil AR and Yadav DK: Comparative standardization study of three triphala churna formulation. Int. J. Pharmacog 2017; 4(2): 71-78.
17. Shivakumar A, Paramashivaiah S, Surappa R, Anjaneya, Hussain J and Ramachandran S: Pharmacognostic Evaluation of Triphala Herbs and Establishment of Chemical Stability of Triphala Caplets. Int. J. Pharm. Sci. Res. 2016; 7: 244-251.
18. Sangolgi B, Simpi P, Burigi P and Joshi D: Pharmasuitical study of thriphala arka- a research article. Int. J. Curr. Med. Sci. 2017; 7: 257-262.
19. Bahuguna Y, Zaidi S, Kumar N and Rawat K: Standardization of Polyherbal Marketed Formulation Triphala Churna. RRJPP 2014; 2: 28-35.
20. Biradar YS, Sharma P and Khandelwal KR: Preparation, method of optimization and physicochemical evaluation of traditional formulation, Triphala Mashi. Ind. J. Trad. Knowledge 2007; 6: 292-297.
21. Kaushika R, Kumara P, Rathorea V and Roy GS: Physicochemical evaluation of Triphala churna. Int. J. Pharma. Sci. 2015; 1; 71-74.
22. Jadhav RP: Regional variation of tannin in triphala drugs and triphala churna. IJRBAT 2017; 2: 1-3.

How to cite this article:
Kondalkar A, Kondalkar SA, Kumar V, Mangal AK and Subhose V: Effect of proportion composition variation on physicochemical parameters of Triphala. Int J Pharm Sci & Res 2018; 9(10): 4280-85. doi: 10.13040/IJPSR.0975-8232.9(10).4280-85.