J Young Pharm, 2021; 13(3) : 226-229
A multifaceted peer reviewed journal in the field of Pharmacy
www.jyoungpharm.org | www.phcog.net

Original Article

Comparative Quality Evaluation of Different Brands of '*Triphala*': A Polyherbal Medicine in Sri Lanka

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ABSTRACT

Background: *Triphala* is a well-known polyherbal formulation widely used as an herbal, complementary and alternative medicine for various ailments, especially for gastrointestinal problems, and it is used as a dietary supplement for long years. **Objectives:** Objective of this study was to comparatively evaluate the quality of three marketed brands of *Triphala* tablets (Brand T_1 and T_2) and capsules (Brand T_3) which are available in Sri Lanka. **Materials and Methods:** This laboratory based experimental qualitative study was done among three brands of *Triphala*. Phytochemical screening was carried out for all those brands. Physicochemical tests were also performed to determine pH, moisture content, and loss on ignition, ash values and extractive values. Data were analysed by using Statistical Package of Social Sciences 23. **Results:** Physicochemical tests revealed that all three brands failed to comply with pharmacopoeial limits at least in one physicochemical test. According to ANOVA test, there was significant difference between results of physicochemical tests of marketed brands of *Triphala*. **Conclusion:** All three *Triphala* brands did not meet the quality standards. Quality of herbal medicine marketed in Sri Lanka should be monitored and regularized by relevant authorities.

Key words: Marketed Brands, Physicochemical Tests, Phytochemical Screening, Quality, Sri Lanka, *Triphala*.

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DOI: 10.5530/jyp.2021.13.46

INTRODUCTION

The World Health Organization has been estimated that 80% of the population in developing countries depends on traditional herbal medicines. The herbal products are widely used as a complementary and alternative medicine for different ailments,[1-3] and also people use those as dietary supplements[4,5] in Sri Lanka.

Triphala is a poly-herbal medicine, which consists of dried fruits of three medicinal plants which are *Terminalia chebula* Retz (Family Combretaceae), *Terminalia bellerica* Roxb (Family Combretaceae) and *Phyllanthus emblica* L. (Family Phyllanthaceae) in equal proportions, based on Ayurvedic Formulary of India (AFI). Commonly, *Triphala* is used for treating a variety of ailments, especially for gastrointestinal problems[6,7] and used for prevention of dental caries[8-10] and cardio-cerebral vascular diseases.[11] A randomized controlled trial which was done to test the effects of *Triphala* formulation, on serum lipid parameters in patients receiving atorvastatin at Sri Lanka stated that the use of that *Triphala* as single therapy for lipid regulation.[12]

Based on scientific studies related to the *Triphala*, it possesses anti-inflammatory,[13,14] anti-microbial,[15] anti-arthritic,[14,16] antioxidant,[17] analgesic, rejuvenating (anti-ageing),[1,8,18] hypoglycaemic[19] and anticancer properties.[20,21] It also possesses chemo-protective, immune-modulating, anti-mutagenic, adaptogenic, and radio-protective effects.[8-10]

Pharmacognostic techniques used in the standardization of plant material include morphological features, physicochemical characteristics, fluorescence analysis and preliminary phytochemical analysis.[18,22] These standards are of utmost importance in finding out genuinely and in the detection of adulterants in marketed drug.[23]

Nowadays, most of the herbal medicines are produced as tablets or capsules. *Triphala* is well known poly herbal medicine, and it is used in the treatment of different ailments in Sri Lanka. Therefore, this study was done to comparatively evaluate the mostly available three *Triphala* brands in the form of tablets and capsules in Sri Lanka.

MATERIALS AND METHODS

The study was limited to tablets and capsules of *Triphala*. Three mostly available brands of *Triphala* tablets (T_1 and T_2) and capsules (T_3) in Sri Lanka were included in this study. Description of these three brands were given in Table 1. Powder sample that were obtained from these brands, was used for preliminary phytochemical screening and physico-chemical analysis. All tests were carried out for the three brands within their expiry date.

Preliminary phytochemical screening

Preliminary phytochemical screening of aqueous extracts of each brand of *Triphala* powder was performed to detect various bioactive substances such as alkaloids, carbohydrates, reducing sugar, flavonoids, saponins, tannins, steroids, proteins, amino acids, glycosides, phenols, terpenoids and ascorbic acid.[3,24]

Physicochemical analysis

Each test was done according to standard methods prescribed in World Health Organization guidelines for quality control methods for herbal materials, Association of Official Agricultural Chemists International methods and Indian Pharmacopeia and the Ayurvedic Pharmacopeia.[25-27]

Table 1: Details of *Triphala* products.

Code	Manufacturing date	Expiry date	Batch number	Dosage form
T_1	01/07/2020	01/07/2021	031054	Tablet
T_2	29/01/2020	28/01/2022	B 107	Tablet
T_3	03/12/2019	02/12/2021	698 A	Capsule

Table 2: Phytochemical screening of Different Brands of *Triphala*.

Phytochemicals	Test	T_1	T_2	T_3
Alkaloids	Mayer's test	-	-	-
	Wagner's test	-	-	-
Carbohydrates	Molisch's test	+	+	+
Reducing sugar	Fehling's test	+	+	+
Flavonoids	Alkaline reagent test	+	+	+
	Lead acetate test	+	+	+
Saponins	Foam test	+	+	+
Tannins	Braymer's test	+	+	+
Steroids	Salkowski's test	+	+	+
Proteins	Millon's test	-	-	-
Amino acids	Ninhydrin test	-	-	-
Glycosides	Keller Killiani's test	+	+	-
Phenols	$FeCl_3$ test	+	+	+
	Lead acetate test	+	+	+
Terpenoids	Copper acetate test	-	-	-
Ascorbic acid	Vitamin C test	+	+	+

+ Present; - Absent

Physicochemical tests such as determination of pH value,[3] moisture content (Loss on Drying), Loss on Ignition,[28] ash values (Total ash, Acid-insoluble ash, Water-soluble ash) and extractive values (Ethanol-soluble and Water-soluble extractive value) were done in triplicate for each brands.

Statistical analysis

Statistical analysis was performed using the statistical package of social science (SPSS) 23 and results were presented as mean values and standard deviations. One-way ANOVA (analysis of variance) test was used to compare results of physicochemical tests of different brands. *P* value less than 0.05 will be considered as statistically significance difference.

RESULTS

In this qualitative experimental study, the three different marketed brands were comparatively evaluated using physicochemical and phytochemical analysis.

Phytochemical screening

Based on the Table 2, Phytochemical screening of aqueous extracts revealed that carbohydrates, reducing sugar, flavonoids, saponins, tannins, steroids, phenols and ascorbic acid were present in all the brands while glycosides was present only in T_1 and T_2. Alkaloids, proteins, amino acids and terpenoids were absent in all brands.

Table 3: Physicochemical Parameters of Different Brands of *Triphala*.

Parameter (%)	Brand T_1 (M±SD)	Brand T_2 (M±SD)	Brand T_3 (M±SD)	Standard (IP)
pH	02.83±0.025	02.87±0.012	03.40±0.015	No limit
Loss on Drying	07.21±0.026	08.82±0.079	10.09±0.195	NMT 12.00
Loss on Ignition	95.97±0.207	96.56±0.006	94.73±0.025	No limit
Total Ash Value	03.65±0.018	03.17±0.021	04.22±0.037	NMT 8.00
Acid-Insoluble Ash Value	04.11±0.103	04.44±0.027	02.56±0.057	NMT 3.00
Water-Soluble Ash Value	03.55±0.051	02.88±0.018	03.78±0.037	No limit
Ethanol-Soluble Extractive Value	32.21±0.634	28.33±0.273	31.36±2.594	NLT 25.00
Water-Soluble Extractive Value	49.34±0.792	39.32±0.139	17.73±0.323	NLT 35.00

M: Mean; SD: Standard Deviation; NMT: Not more than; NLT: Not less than

Physicochemical analysis

According to the Table 3, Brand T_3 had higher values in pH, loss on drying, total ash and water-soluble ash values than other two (T_1 and T_2) brands. Brand T_2 had higher loss on ignition and Acid-insoluble ash values, while T_1 had higher ethanol-soluble and water-soluble extractive values. All physicochemical parameters except acid-insoluble ash value in T_1 and T_2 and water-soluble extractive value in T_3 were complied with the Standard limits mentioned in Indian Pharmacopoeia. The mean comparison of all three brands by using one-way Analysis of variance (ANOVA) statistical test showed significant difference in physicochemical parameters of all tested brands (< 0.05) except in ethanol-soluble extractive value in which no significant difference (Between T_1 and T_3 and T_2 and T_3) was observed (>0.05).

DISCUSSION

Quality of three brands of *Triphala* was evaluated using phytochemical screening and physicochemical tests.

Several studies on preliminary phytochemical screening revealed that the aqueous and alcohol extracts of *Triphala* powder consisted of phenolics, tannins, ascorbic acid, flavonoids, sugar, starch and fixed oil.[18,21-23] Generally, polyphenols, tannins, ascorbic acid, flavonoids, sugar and starch are major phytoconstituents in *Triphala*.[2,29-32] In this study, phytochemical screening of aqueous extract confirmed the presence of carbohydrates, reducing sugar, flavonoids, saponins, tannins, steroids, phenols and ascorbic acid in all three brands while glycosides were present only in brands T_1 and T_2. Alkaloids, proteins, amino acids and terpenoids were absent in all three brands. In the previous study on *Triphala churna*, the phytochemical screening of the aqueous extract of different brands revealed the presence of carbohydrates, steroids, flavonoids, tannins, saponins, phenols and ascorbic acid while glycosides only

in one brand with the absence of alkaloids, proteins, amino acids and terpenoids.[3] However, another study done for a marketed brand and a lab-prepared sample confirmed the presence of alkaloids and the absence of saponins and proteins.[30] Another study confirmed the absence of glycosides, proteins, saponins, alkaloids and phytosterols.[29] Current and previous studies showed variations in the phytochemical constituents. Since phytochemical screening is a qualitative study, it may not detect some phytoconstituents which was in the low quantity in the samples.

The African Pharmacopeia stated that physicochemical evaluation of drugs is an essential parameter in detecting adulteration or improper handling of drugs.[33] In this study, the pH of the all *Triphala* brands was in the range of 2.83 to 3.40. Presence of ascorbic acid may contribute to the low pH of the *Triphala* formulations. Previous study done with different brands of *Triphala* churna in India also showed comparable pH values[3] while a study reported different pH value.[34] Loss on drying was used to determine the volatile constituents in powdered drugs. Low moisture content is often desirable for stability of herbal drugs. Low moisture content of drugs could prevent microbial growth during storage.[35-37] In this study, all three brands were within the limit (≥ 12% w/w) in loss on drying values. However, T_3 had high value of loss on drying compare to others. Even though loss on ignition is one of the most widely used methods for determining the content of organic matter in herbs, it was not included as standard tests. In this study, the loss on ignition values of T_1, T_2 and T_3 were comparable with the previous study done in India.[3]

Ash values were used to determine the quality and purity of the unsophisticated drug.[35] Low ash values indicate more purity and quality. High ash values may be due to contamination, substitution, adulteration or carelessness in preparing the herbal formulations. Acid-insoluble ash value indicates presence of siliceous impurities. Water-soluble ash value is due to presence of inorganic contents. In this study, ash values were within the standard limit (≥ 8%). These values were comparable with the findings of previous studies.[3,30] Acid insoluble ash values of 03 brands were in the range from 2.56 to 4.44% w/w. The acid insoluble ash value of T_3 was within the standard limit (IP limit of ≥ 3.0%) while T_1 and T_2 were exceed the limit. The acid-insoluble ash value of the T_3 was comparable with the previous study.[3] High acid-insoluble ash value was observed in two brands in this study, and it might be due to the presence of more earthy materials. The water-soluble ash values of the *Triphala* brands ranged from 2.88 to 3.78% w/w. But these values were not comparable with water-soluble ash values of previous studies.[3,21,22]

The extractive values (alcohol and aqueous) are useful to estimate the soluble chemical constituents in the crude drug.[36] Low Ethanol soluble extractive values indicate usage of exhausted material, adulteration, using less amount of herbals, incorrect processing or storage. In this study, the values of all three brands were within the IP limit (≤ 25.0%) and could be comparable with the previous study.[3] Water-soluble extractive value of T_3 was within standard limit. But T_1 and T_2 were exceed the limit (≥35.0%) and same results was reported in the previous studies.[3,29,30,34] In the present study, water-soluble extractive value was higher than alcohol extractives. It could be due to that most of the phytoconstituents in the sample were solubilised in water. Additions of exhausted material, adulteration or incorrect processing might contribute to less water extractive values. In this study, all three brands failed to satisfy the standard limit in at least one physicochemical test. ANOVA test also revealed that statistically significant differences (<0.05) in all physicochemical tests except in ethanol soluble extractive value.

Difference in the results of phytochemical screening is due to several reasons such as preparation procedure, storage method, seasonal variations, and collection of plant materials from different phytogeographical regions, variation in quantity of each plant, adulteration or substitution. Since different excipients are added for ease of production of tablets and capsules, they may have an impact on quality control tests such as physicochemical tests and phytochemical screening. Currently, many herbal formulations are processed in to capsules and tablets. So that existing standard limits should be revised for herbal based tablets and capsules.

CONCLUSION

There was a significant difference in the most physicochemical tests among all three marketed *Triphala* brands. All three brands failed to comply with standard limit in at least one physicochemical test. Herbal based drugs should be monitored for their quality by relevant authorities. Further, standard limits used for assessing the quality of herbal base capsules and tablets should be revised.

ACKNOWLEDGEMENT

The authors acknowledge University of Jaffna to support this study.

CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

ABBREVIATIONS

AFI: Ayurvedic Formulary of India; **WHO:** World Health Organization; **SPSS:** Statistical Package of Social Science; **ANOVA:** Analysis of Variance; **M:** Mean; **SD:** Standard Deviation; **NMT:** Not more than; **NLT:** Not less than.

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Article History: Received: 28-04-21;Revised: 21-05-21;Accepted: 15-06-21
Cite this article: Premathilake R, Thuvaragan S, Sanmugarajah V, Srikaran R, Krishnapillai N. Comparative Quality Evaluation of Different Brands of '*Triphala*': A Polyherbal Medicine in Sri Lanka. J Young Pharm. 2021;13(3):226-9.