

Pharmacognostical Standardization, Physico-Chemical Analysis, And Antioxidant Activity of Shwadamshtadi Churna

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ABSTRACT

Shwadamshtadi churna is a traditional ayurvedic formulation widely utilised for its extensive therapeutic applications, especially in inflammatory and metabolic disorders. The scientific evaluation of phytochemical characteristics and antioxidant potential is necessary to ensure consistency, quality control, and standardization. This paper aims to provide data on the preparation, authentication, analysis, and validation of the antioxidant activity of Shwadamshtadi Churna. The raw drugs were authenticated, and the churna was prepared following standard guidelines and procedures. It was evaluated for pharmacognostic and physicochemical parameters to identify major phytoconstituents, microbial contamination, and antioxidant activity assessment using 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging assay. The physicochemical parameters results were as follows: 8.581% moisture content, 7.593% ash value, 1.687% acid-insoluble ash, 21.782% water-soluble extractive, 15.680% alcohol soluble extractive, and absence of pathogenic organisms, indicating no microbial contamination. The churna demonstrated concentration-dependent DPPH radical scavenging activity, with 62.5 µg/ml showing maximum inhibition of 96.89%, indicating robust antioxidant potential. The formulation fulfilled all the mandatory physico-chemical parameters of the Ayurvedic Pharmacopoeia of India, indicating rigor in the production and processing. The extractive values demonstrate optimal solubility of the bioactive constituents. Numerous studies are available to substantiate the antioxidant, anti-inflammatory, and cytoprotective effects of amalaki, guduchi, and gokshura. The results of the DPPH assay reinforce the synergistic antioxidant effect, hence proving the rasayana potential. This study establishes comprehensive quality control parameters for the preparation and analysis of Shwadamshtadi churna with its rich phytochemical profile. It also validates the antioxidant activity, providing a scientific basis for its therapeutic utility in oxidative and inflammatory pathologies. This study also provides a easily reproducible framework for consistent production and analysis of Shwadamshtadi churna for maximum therapeutic efficacy.

Keywords: Antioxidant activity, DPPH, Phytochemical analysis, Shwadamshtadi churna, Standardization..

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INTRODUCTION

Ayurveda, the Indian system of medicine, is one of the most renowned, widely accepted traditional systems of medicine based on scientific fundamentals¹. It provides various therapeutic solutions to ailments through formulations and procedures. Polyherbal formulations form the backbone of ayurvedic pharmaco-therapeutics². *Shwadamshtadi churna* is made with easily available drugs and administered in the form of *churna* (powder). *Churna* preparations are versatile and allow rapid onset of action due to their fine powder form, which enhances dissolution and absorption. *Shwadamshtadi churna* is known for its rejuvenative, immunomodulatory, and antioxidant

properties³. The World Health Organisation emphasizes the need for scientific, robust standardization of ayurvedic formulations for quality control and reproducibility⁴. Oxidative stress is known to significantly contribute to the pathogenesis of cardiovascular, neurodegenerative, and immune disorders⁵. Herbs serve as a natural source of antioxidants and are proven to have better safety, efficacy, and multi-target therapeutic potentials compared to synthetic alternatives⁶. Ayurvedic formulations with a rich phytochemical repository and antioxidant properties contribute to the therapeutic efficacy⁷.

Although literature exists on the therapeutic potential of this *churna* in the form of advanced docking studies, there

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emerges a need for comprehensive documentation, pharmacognostical standardization, and evaluation of the biological activity of *Shwadamshtadi churna* to ensure ease in production and administration.

The present study was hence undertaken to:

Establish standardization of pharmacognostical parameters according to API guidelines

Evaluation of microbial contamination to warrant safety

Assess the antioxidant potential through the DPPH radical scavenging assay.

This study aims to provide scientific validation and provide reference standards for quality control of *Shwadamshtadi Churna*

MATERIALS AND METHODS

Preparation of Shwadamshtadi Churna:

The formulation was prepared as per Ayurvedic methodology at GMP-certified KLE Ayurveda Pharmacy, Khasbag, Belagavi. The raw materials were procured from authentic sources, subjected to authentication in the AYUSH-approved Drug Testing Laboratory, Central Research Facility, CRF. The dried individual ingredients were cleaned and pulverised separately using a mechanical grinder. The obtained powdered ingredients were sieved (80 mesh size) to ensure uniformity in particle size. The powders were then mixed in equal quantities thoroughly, and a homogenous *Shwadamshtadi churna* was obtained. The ingredients, part used, and yield are depicted in Table 1.

Table 1: Actual output obtained from the individual ingredients.

S.No	Ingredients	Part used & quantity	Yield
1.	<i>Guduchi-Tinospora cordifolia</i> (Thunb.) Miers	Stem-10 Kg	8.120
2.	<i>Gokshura-Tribulus terrestris</i> Linn.	Fruit-10 Kg	6.840
3.	<i>Amalaki-Emblica officinalis</i> Gaertn.	Fruit-10 Kg	10.040



Authentication & Quality Control Testing:

The prepared churna was submitted to the Central Research Facility, Shri B M Kankanawadi Ayurveda Mahavidyalaya, Shahapur, Belagavi (AYUSH Approved ASU Drug Testing Laboratory, License No. TL-8/2011) for organoleptic & physicochemical evaluation according to the standard guidelines in the Ayurvedic Pharmacopoeia of India (API)⁸.

Organoleptic characteristics such as form, color, taste, and odour were evaluated. Physicochemical parameters were determined as follows:

Moisture content was determined by the loss on drying method at 105°C until a constant weight is attained. Total ash was determined by incineration at 450°C in a muffle furnace. Acid-insoluble ash was determined by the ash residue insoluble in dilute hydrochloric acid. Water-soluble extractive by extractable matter in water using the cold maceration method, and Alcohol-soluble extractive by extractable matter in alcohol (ethanol) using the cold maceration method.

All determinations were performed in triplicate and results expressed as percentage (% w/w). Table 2 shows the testing of individual ingredients of *Shwadamshtadi churna*.

Table 2: Organoleptic and physicochemical evaluation of individual ingredients of *Shwadamshtadi churna*

Drug name	Macroscopic features	Foreign matter (%)	Ash value (%)	Acid insoluble ash (%)	Water soluble extractive (%)	Alcohol soluble extractive (%)	API Limits	Interpretation
<i>Gokshura</i>	Light green, sweet-astringent, unpleasant odor	0.618	14.419	3.160	14.385	4.328	FM ≤2, Ash ≤17, AIA ≤4, WSE ≥12, ASE ≥2	All within limits, standard quality
<i>Amalaki</i>	Grey-black, sour-astringent	Nil	3.073	0.694	52.641	43.062	FM ≤3, Ash ≤7, AIA ≤2, WSE ≥50, ASE ≥40	All within limits, high quality
<i>Guduchi</i>	Light brown, bitter	Nil	6.594	0.738	13.408	4.301	FM ≤2, Ash ≤16, AIA ≤3, WSE ≥11, ASE ≥3	All within limits, standard quality

Abbreviations: FM = Foreign Matter, AIA = Acid Insoluble Ash, WSE = Water Soluble Extractive, ASE = Alcohol Soluble Extractive.

Qualitative and quantitative test for microbial contamination for the detection and quantification of total microbial load was performed as per API guidelines as follows:

Specific microorganisms (qualitative): Testing for *Escherichia coli*, *Staphylococcus aureus*, *Pseudomonas aeruginosa*, and *Salmonella abony* using standard culture media.

Microbial limit tests (quantitative): Total bacterial count and total fungal count enumeration using the plate count method.

Evaluation of antioxidant activity by DPPH Radical Scavenging Assay : The antioxidant activity of the alcoholic extract of *Shwadamshtadi churna* was evaluated using the DPPH (2,2-diphenyl-1-picrylhydrazyl) radical scavenging method^{9,10}.

Preparation of Extract: 10g of *Shwadamshtadi churna* was macerated with 100 ml of ethanol for 24 hours and subjected to intermittent shaking. The extract was obtained by filtration and concentration.

DPPH assay: the DPPH solution 0.1mM was prepared in methanol. Varied concentrations of the alcoholic extract (15.625, 31.25, 62.5, 125, 250, and 500 µg/ml) were

prepared. 1 ml of each concentration was mixed with 3 ml of DPPH solution and incubated at room temperature for 30 minutes. A UV-visible spectrophotometer was utilised to measure the absorbance at 517 nm against a blank.

Calculation: the percentage of radical scavenging activity was calculated using the formula:

$$\text{Radical Scavenging Activity (\%)} = \frac{A_{\text{control}} - A_{\text{sample}}}{A_{\text{control}}} \times 100$$

where A_{control} is the absorbance of the control (DPPH solution without sample) and A_{sample} is the absorbance of the test sample with DPPH solution.

IC₅₀ Determination: The IC₅₀ value (concentration required to scavenge 50% of DPPH radicals) was determined by plotting concentration versus percentage radical scavenging activity.

All experiments were performed in triplicate.

RESULTS

Organoleptic Characteristics:

Table 3 presents the organoleptic evaluation of *Shwadamshtadi Churna*.

Table 3: Results of Organoleptic parameters of *Shwadamshtradi churna*

Parameter	Observation
Form	Churna (fine powder)
Colour	Creamish
Taste	Slightly bitter
Odour	Characteristic

The formulation was a fine, free-flowing powder with a creamish colour, characteristic odour (aromatic), and slightly bitter taste, which was consistent with the nature of constituent herbs as shown in the figure 1.

Figure 1: Final *churna* obtained

Physicochemical Parameters:

Table 4 presents the physicochemical parameters and analysis results.

Table 4: Results of physicochemical parameters

Parameter	Result (% w/w)
Moisture content	8.581
Total ash value	7.593
Acid-insoluble ash	1.687
Water-soluble extractive	21.782
Alcohol-soluble extractive	15.680

Moisture Content: The moisture content of 8.581% indicates adequate drying of the formulation, which is critical for preventing microbial growth and ensuring stability during storage¹¹.

Ash Values: Total ash value of 7.593% represents the total inorganic content. Acid-insoluble ash of 1.687% indicates minimal siliceous matter and earth contamination, confirming proper cleaning and processing of raw materials¹².

Extractive Values: Water-soluble extractive value of 21.782% and alcohol-soluble extractive value of 15.680% indicate the proportion of polar and semi-polar phytochemical constituents, respectively. Higher water-

soluble extractive suggests the presence of substantial hydrophilic compounds, including polysaccharides, glycosides, and phenolic compounds¹³.

These physicochemical parameters are within acceptable limits for Ayurvedic churna formulations and can serve as reference standards for quality control.

Microbial Contamination Assessment

Microbial safety evaluation demonstrated the absence of pathogenic microorganisms and acceptable microbial load, as shown in Table 5

Table 5: Results of microbial contamination assessment

Test	Limit/Standard	Result
Specific Microorganisms (Qualitative)		
<i>Escherichia coli</i>	Absent/100ml	Absent
<i>Staphylococcus aureus</i>	Absent/100ml	Absent
<i>Pseudomonas aeruginosa</i>	Absent/100ml	Absent
<i>Salmonella abony</i>	Absent/100ml	Absent
Microbial Limit Test (Quantitative)		
Total bacterial count	30-300 cfu/ml	No growth
Total fungal count	10-100 cfu/ml	No growth

The absence of pathogenic organisms (*E. coli*, *S. aureus*, *P. aeruginosa*, *S. abony*) and negligible microbial count confirms microbiological safety of the formulation. This meets the safety standards prescribed by API and WHO guidelines for herbal medicines.

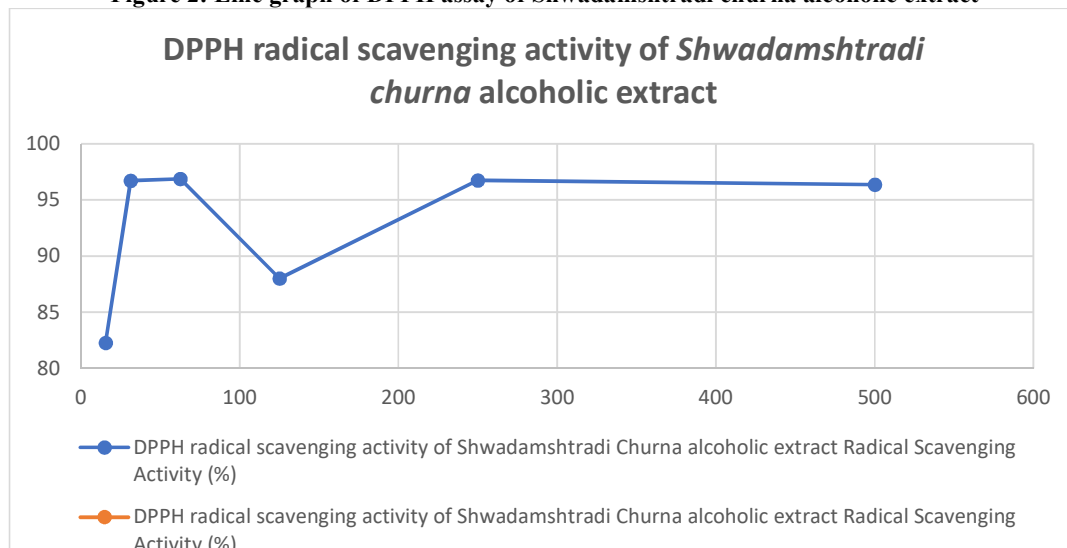
Antioxidant Activity:

The alcoholic extract of Shwadamshtadi Churna demonstrated dose-dependent DPPH radical scavenging activity. Results are presented in Table 6.

Table 6: DPPH radical scavenging activity of Shwadamshtadi Churna alcoholic extract

Concentration ($\mu\text{g/ml}$)	Radical Scavenging Activity (%)
15.625	82.28
31.25	96.72
62.5	96.89
125	88.00
250	96.76
500	96.38

Figure 2: Line graph of DPPH assay of Shwadamshtadi churna alcoholic extract



The formulation exhibited strong antioxidant activity across all tested concentrations. Maximum radical scavenging activity of 96.89% was observed at 62.5 $\mu\text{g/ml}$ concentration. Even at the lowest concentration tested (15.625 $\mu\text{g/ml}$), the extract demonstrated substantial activity of 82.28%, indicating potent antioxidant constituents.

The dose-response relationship showed that activity increased from 82.28% to 96.89% with concentration increase from 15.625 to 62.5 $\mu\text{g/ml}$. Beyond this concentration, the activity plateaued, maintaining values above 88% at all higher concentrations (125-500 $\mu\text{g/ml}$).

This plateau pattern suggests saturation of radical scavenging at moderate concentrations.

The IC_{50} value (concentration required to achieve 50% radical scavenging) is 15.625, which appears to be well below the lowest concentration tested, indicating very high antioxidant potency of the formulation. All results represent the mean of three independent readings, confirming the reproducibility of the antioxidant effect.

DISCUSSION

The present study provides standardized data for improving the ease in the preparation, analysis, and antioxidant

potential of *Shwadamshtadi churna*. Traditional preparation methods & pharmaceutical processes, coupled with contemporary analytical techniques, provide a solid framework for quality assurance and standardization of this formulation.

The constituent herbs of this formulation (*Guduchi*, *Gokshura* & *Amalaki*) impart the distinct organoleptic characters of creamish colour, slightly bitter taste, and characteristic odour. These subjective parameters provide data on the initial quality indicators as they are observable readily, and abnormalities in the above can indicate adulteration or deterioration.

The physicochemical parameters are well within API standards for ayurvedic churna formulations¹⁴. The moisture content of 8.581% indicates powder flowability and is appropriate for the prevention of microbial proliferation. API standards postulate that moisture content below 10% is acceptable for churna formulations, as excessive moisture is known to lead to caking, microbial growth, and loss of active constituents¹⁵. The ash values indicate proper processing, and the extractive values suggest the presence of substantial bioactive phytochemicals.

The higher water-soluble extractive (21.782%) compared to alcohol-soluble extractive (15.680%) indicates the predominance of hydrophilic constituents like polysaccharides, glycosides, saponins, tannins, and phenolic compounds¹⁶ that significantly contribute to therapeutic efficacy. The extractive values also serve as quantitative standards for the assessment of the efficiency of extraction and concentration of active principles in the batch prepared.

The quality of the formulation for pharmaceutical application is confirmed by microbial safety assessment. The absence of pathogenic organisms and negligible microbial load shows compliance with safety regulatory standards¹⁷.

The strong antioxidant potential assessed by DPPH radical scavenging activity provides scientific validation for therapeutic efficacy. High antioxidant activity at low concentrations (82.28% at 15.625 µg/ml) indicates the potent phytochemicals responsible for antioxidant activity. The plateau effect at concentrations above 62.5 µg/ml suggests that the *churna* exhibits maximal radical scavenging at moderate doses. The DPPH assay can be utilised as a robust, first-line assay for radical scavenging owing to its simplicity, reproducibility, and provides a strong global index for radical scavenging¹⁸.

The antioxidant activity can be attributed to the presence of varied phytochemical constituents, namely phenolic compounds, flavonoids, tannins, alkaloids, and terpenoids¹⁹. Extensive data is available on the presence of tannins, glycosides, alkaloids, and carbohydrates in *Amalaki*, alkaloids, steroids, and glycosides in *Guduchi*, and saponins, flavonoids, glycosides, alkaloids, and amino acids in *Gokshura*²⁰. These compounds act through multiple mechanisms, such as electron transfer, metal ion chelation, radical stabilization, and hydrogen donation²¹. The DPPH assay especially measures the hydrogen-donating capacity, which is a primary antioxidant mechanism.

The pathogenesis of various chronic diseases is influenced by oxidative stress. Excess free radicals lead to oxidative damage to DNA, lipids, and proteins, leading to the progression of disease²¹.

Herbs are a potent source of antioxidants that are capable of neutralising free radicals and modulating redox-sensitive signaling pathways naturally²². The DPPH assay proves the strong antioxidant activity of *Shwadamshtadi churna*, supporting its therapeutic application in conditions associated with oxidative stress.

DPPH assays of several ayurvedic formulations, like *Arogyavardhini vati* and *Amalakayas rasayana*, demonstrated significant antioxidant activity, providing substantial support to their therapeutic efficacy^{23,24}. The consistently high activity (>82%) across all concentrations in the present study of *Shwadamshtadi churna* places it among the highly potent formulations.

This research integrates traditional knowledge with contemporary pharmaceutical sciences, providing scientific evidence to the traditional thought process. It provides a wider acceptance and approach to the utilisation of Ayurvedic formulations.

The quality control parameters established through this study can serve as reference standards for the manufacturing, testing, and consistent production of quality churna. These standards provide, authentication of genuine *churna*, detection of adulteration or substitution, consistency in prepared batches and quality parameters and relation to therapeutic efficacy.

CONCLUSION

The present study establishes a scientific foundation and provides comprehensive standardisation parameters of *Shwadamshtadi churna*, including organoleptic characteristics, physicochemical constituents, and microbial safety limits. The formulation demonstrates potent antioxidant activity with the DPPH radical scavenging assay, exceeding 82% across all tested concentrations, reaching maximum activity of 96.89% at 62.5 µg/ml. The traditional use of *Shwadamshtadi churna* is scientifically validated, providing reference standards for standardisation and quality control. Research on detailed phytochemical profiling using advanced techniques for the identification of biomarkers is also available that further establishes and clinically validates the therapeutic potential of *Shwadamshtadi churna*. This paper provides data for improving the ease of production and standardisation, in turn promoting the widespread utilisation of *Shwadamshtadi churna*.

CONFLICT OF INTEREST

The authors declare no conflict of interest in this research.

Limitations

Further investigation into the phytochemical profile with advanced techniques like High Performance Liquid Chromatography (HPLC), Gas Chromatography-Mass Spectrometry (GC-MS), and Liquid Chromatography-Mass Spectrometry (LC-MS) will provide specificity in the bioactive constituents to, in turn, validate its therapeutic potential.

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Declaration of generative AI and AI-assisted technologies in the manuscript preparation process.

Statement: During the preparation of this work, the author(s) used Claude AI & Perplexity AI to identify and condense indexed published articles, for rephrasing text to enhance scientific tone and to correct minor spelling errors. After using this tool/service, the author(s) reviewed and edited the content as needed and take(s) full responsibility for the content of the published article. No AI tools were used for data generation, analysis, interpretation, or the development of scientific content; all substantive material was produced by the authors through independent research

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