RESEARCH PAPER

Formulation, Evaluation and Elemental Standardization of Polyherbal Face Pack Validated by Atomic Absorption Spectroscopy Method

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ABSTRACT

Introduction: Herbal face packs are widely used for skincare due to their natural origin, safety, and therapeutic benefits. The present study aimed to formulate, evaluate, and standardize a polyherbal face pack and determine its elemental content using a validated Atomic Absorption Spectroscopy (AAS) method. **Materials and Methods:** The face pack was prepared using Multani Mitti, Red Lentil Powder, Gram Flour, Lodhra Powder, Turmeric Powder, Orange Peel Powder, and Aloe Vera Powder. Three formulations (F1, F2, and F3) were developed and evaluated for organoleptic and physicochemical characteristics. The AAS method was validated according to ICH guidelines for linearity, accuracy, precision, specificity, robustness, LOD, and LOQ.**Results**: A Atomic Absorption Spectroscopy (AAS) method was successfully validated (high linearity, precision %RSD < 2%, and robust recoveries) to quantify heavy metal ions including zinc, iron, copper, and manganese. Skin-compatible pH of 5.34 and satisfactory elemental profile.**Conclusion**: The analysis confirmed that all elemental levels in the face pack are well within safe cosmetic thresholds, establishing a scientifically validated, non-toxic, and cost-effective herbal skincare product. The developed polyherbal face pack was found to be safe, effective, economical, and suitable for skincare applications.

KEYWORDS: Polyherbal Face Pack, Atomic Absorption Spectroscopy (AAS), Method Validation, Elemental levels, Iron, Zinc, Copper, Manganese, Safe Cosmetic Thresholds, Skin Care product.

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1. INTRODUCTION

The largest organ in the human body is the skin which acts as the primary protective barrier. against external elements like dust, toxins, microbes, and dangerous radiation. Additionally, it is crucial for controlling bodily temperature, feeling, and general homeostasis. [1] In addition to providing physical protection, healthy skin is crucial for confidence and aesthetic appeal. However, several skin issues, including acne, pigmentation, dryness, dullness, and premature aging, have become more prevalent as a result of modern lifestyle changes, stress, poor eating habits, and ongoing exposure to environmental contaminants.[2] Natural and herbal cosmetics have been increasingly popular recently. Compared with synthetic cosmetics, herbal cosmetics—which are made with plant-based ingredients—are thought to be safer, more environmentally friendly, and more skin-friendly. Although synthetic cosmetics might produce immediate results, they can also cause adverse effects such as

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allergies, skin irritation, and long-term harm. [3,4] Herbal formulations, on the other hand, are typically non-toxic, gentle, and suitable for long-term use. Ayurveda and other traditional medical systems have long highlighted the value of natural substances for preserving skin health. [4,5] Mukhalepa, the application herbal paste to the face, is one such custom. Many people use herbal face packs to cleanse, nourish, revitalize, and improve the skin's general texture. Usually applied as a paste, these treatments help remove impurities, tighten the skin, and improve blood circulation by forming a thin layer on the skin that dries after application. A variety of natural substances are combined to create herbal face packs, each of which adds unique health benefits. [6,7,8] Multani mitti, red lentil powder, gram flour, lodhra powder, turmeric, orange peel powder, and aloe vera make up the formulation used in this study. Multani mitti is renowned for its superior deep-cleansing and oil-absorbing qualities, which enable it to effectively remove excess sebum and pollutants from the skin. As a natural exfoliating agent, red lentil powder helps to improve the texture of the skin and eliminate dead skin cells. [6,7,9] Gram flour's cleaning and complexion-enhancing qualities make it a popular ingredient in traditional skincare. [6,7,10] Lodhra powder is well known for its healing and skin-tightening properties, which assist in lowering inflammation and enhancing skin tone. Strong anti-inflammatory, antibacterial, and antioxidant qualities make turmeric a popular herbal ingredient that helps manage acne and prevent infections. [11-13] Vitamin C and natural antioxidants included in orange peel powder aid in lightening skin, lessening pigmentation, and creating a more balanced complexion. [14] Aloe vera is well-known for its calming, hydrating, and restorative qualities, which support skin moisture and lessen irritation. These organic components come together to create a mixture that offers several advantages, including skin protection, cleansing, exfoliating, and nourishment. [15] Herbal face packs are a popular option for skincare since they are affordable, simple to make, and appropriate for frequent usage. Ensuring the safety and quality of herbal cosmetics is just as crucial as formulation and evaluation. Despite their advantages, natural substances may contain trace levels of metal ions due to environmental contamination during production, processing, or storage. Excessive levels of some metal ions can be detrimental to the skin and general health. Analyzing and measuring these ions in the formulation is therefore crucial.Atomic Absorption Spectroscopy (AAS) is one of the most used and dependable analytical methods for estimating metal ions. The idea behind this method is that light is absorbed by free atoms in the gaseous state. Because each element absorbs light at a particular wavelength, the presence and concentration of metal ions can be accurately determined. When it comes to identifying trace elements, AAS is renowned for its great sensitivity, accuracy, and specificity. An essential stage in ensuring the accuracy and dependability of the findings is the validation of the analytical method. Method validation includes parameters such as accuracy, precision, linearity, and reproducibility. The estimate of ions in the herbal formulation is guaranteed to be precise and reliable by an approved AAS method. It is possible to ascertain whether the levels of metal ions in the formulation are within safe and acceptable bounds by estimating the ions using a validated atomic absorption spectroscopy method. [16-18] This guarantees that the herbal face pack is safe to apply topically and that it is effective. Therefore, the current study focuses on the formulation and evaluation of an herbal face pack with specific natural constituents as well as the measurement of ionic content using a validated Atomic Absorption Spectroscopy technique. The goal of the study is to create an herbal cosmetic product that is safe, efficient, and affordable while guaranteeing its quality and safety through scientific examination.

2. MATERIALS AND METHODS

2.1. Selection of Measurement Condition for Iron, Zinc, Copper and Manganese by AAS

Before the analysis on AAS, parameters were selected to analyze the Fe, Zn, Cu and Mn content in the formulation. The measurement conditions are shown in Table 1.

Table No 1: Measurement conditions for Fe, Zn, Cu and Mn

Parameter	Condition
Instrument	Atomic Absorption Spectrophotometer (Thermo Fisher Scientific)
Type	Air–acetylene
Fuel Flow Rate	Approximately 2.76 L/min
Slit Width	0.3nm
Burner Height	5.0 mm
Burner Angle	0.00
Measurement Mode	Integrate mode

Integration time	3 seconds
Blank Analysis	Blank analyzed after every sample
Accuracy	Two decimal places
Iron Wavelength	~248.3 nm
zinc Wavelength	~213.9nm
Copper Wavelength	~324.8 nm
Manganese Wavelength	~279.5 nm
Lamp Used	Hollow cathode lamp for each element
Result Expression	Recorded directly in ppm

2.2. Preparation of Standard Solution

3. Methods for preparation of stock solution

3.1. Fe, Cu and Mn.

The quantity of iron salt (ferrous sulphate), copper salt (copper sulphate pentahydrate, $CuSO_4 \cdot 5H_2O$) and manganese salt (manganese sulphate monohydrate, $MnSO_4 \cdot H_2O$) was calculated based on its molecular weight to obtain a final concentration of 1 mg/mL (1000 ppm) of iron, copper and Manganese**.** An accurately weighed quantity of iron salt, such as ferrous sulphate, equivalent to 1 mg/mL or 1000 ppm, was taken and dissolved in deionized water. The solution was then transferred into a 50 mL volumetric flask, diluted up to the mark with deionized water, and mixed thoroughly.

Preparation of Working Standards

From the prepared stock solution, aliquots of 0.1,0.2,0.3,0.4 and 0.5ml were pipetted into separate 50 mL volumetric flasks. Each flask was diluted to the mark with deionized water and mixed well. The final standard solutions were prepared at concentrations of 2 ppm, 4 ppm, 6 ppm, 8 ppm, and 10 ppm.

3.2. Zinc

Zinc oxide (equivalent to 100 mg of zinc) was accurately weighed and transferred into a 100 mL volumetric flask. A sufficient quantity of 0.1 N hydrochloric acid (HCl), prepared using deionized water, was added to ensure proper digestion and complete dissolution of zinc oxide. After dissolution, the volume was made up to the mark with distilled water to obtain a zinc stock solution with a final concentration of 1 mg/mL(1000 ppm).

Suitable aliquots were then taken from the prepared stock solution and further diluted with distilled water to prepare standard solutions of 2 ppm, 4 ppm, 6 ppm, 8ppm, and 10 ppm. The prepared standard solutions were analyzed using Atomic Absorption Spectroscopy (AAS).

Preparation of Sample Solution for AAS

Three formulations, namely F1, F2, and F3, were prepared using the selected ingredients and mixed thoroughly to obtain uniform samples. Approximately 1 g of each formulation was accurately weighed and transferred into separate beakers. A suitable volume of 0.1N HNO3 dilute acid was added to each sample. The mixtures were then subjected to sonication for 10–15 minutes to enhance the extraction of metal ions. The resulting solutions were filtered through Whatman filter paper to remove insoluble matter. The clear filtrates were transferred into separate 50 mL volumetric flasks, diluted up to the mark with deionized water, and mixed well. The prepared sample solutions of F1, F2, and F3 were used for the determination of iron, copper, manganese, and zinc by Atomic Absorption Spectroscopy (AAS).

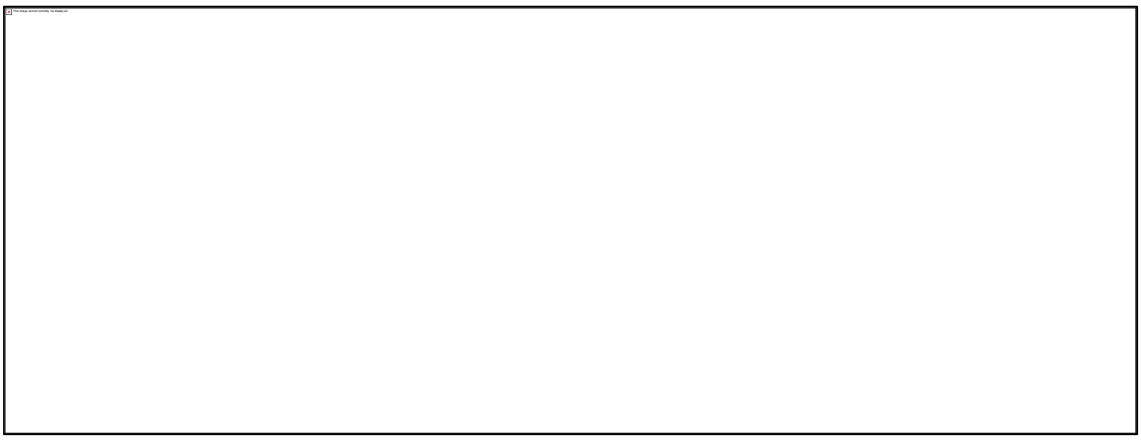

Figure 1 : Schematic diagram of Formulation of face pack

METHOD VALIDATION

Linearity

The concentration of standard solutions of iron, copper, manganese, and zinc was found to be directly proportional to the absorbance within the selected concentration range, indicating compliance with Beer–Lambert's law. Calibration curves were constructed by plotting absorbance versus concentration for each metal, and the corresponding regression equations were obtained. The calibration curves are presented in the respective figures, and the regression data are summarized in the tables. The linearity was evaluated over the concentration range of 2–10 ppm for all four metals. The calibration curves exhibited good linearity with correlation coefficient (R^2) values close to 1, indicating a strong linear relationship between concentration and absorbance.

System Suitability

System suitability was performed to confirm the reliability and reproducibility of the Atomic Absorption Spectroscopy instrument. Standard solutions of zinc, iron, copper, and manganese having concentration of 10 µg/mL were prepared. Each solution was aspirated five times into the instrument and the absorbance values were recorded. The percentage relative standard deviation (%RSD) was calculated for all the elements from the obtained results.

Accuracy

Accuracy of the developed method was determined by standard addition method at three levels (80%, 100%, and 120%) for the estimation of metal ions using Atomic Absorption Spectroscopy (AAS). A pre-analyzed sample solution of each metal ion was spiked with known amounts of standard solution. The analysis was carried out and percentage recovery was calculated.

Formula

% Recovery= Amount found - Original amount\Amount added x 100

Precision

Precision of the Atomic Absorption Spectroscopy method was determined by analyzing standard solutions of zinc, iron, copper, and manganese at a concentration of 10 µg/mL. Each solution was analyzed repeatedly under the same operating conditions and the absorbance values were recorded. The percentage relative standard deviation (%RSD) was calculated to evaluate the precision of the method. Low %RSD values indicated good precision and reproducibility of the analytical method.

Specificity

Specificity of the Atomic Absorption Spectroscopy method was evaluated to determine the ability of the method to accurately measure zinc, iron, copper, and manganese in the presence of other components and possible interferences. Individual standard solutions of each element were prepared and analyzed at their respective wavelengths. The absorbance obtained for each element showed no interference from other elements or reagents, indicating that the method was specific for the determination of zinc, iron, copper, and manganese.

Limit of Quantification (LOQ)

The Limit of Quantification (LOQ) of zinc, iron, copper, and manganese was determined using the calibration curve method in Atomic Absorption Spectroscopy analysis. LOQ represents the lowest concentration of analyte that can be quantitatively estimated with acceptable accuracy and precision. The LOQ was calculated using the following equation:

LOQ = 10 x σ/S or 10 X SD/S

Where:

σ = Standard deviation of the response

S = Slope of calibration curve

The obtained LOQ values indicated the quantification sensitivity of the analytical method for zinc, iron, copper, and manganese.

The Limit of Detection (LOD)

The Limit of Detection (LOD) of zinc, iron, copper, and manganese was determined using the calibration curve method in Atomic Absorption Spectroscopy analysis. LOD represents the lowest concentration of analyte that can be detected but not necessarily quantified

accurately. The LOD was calculated using the following equation:

LOD = 3.3 x σ/S

Where:

σ = Standard deviation of the response

S = Slope of calibration curve

The obtained LOD values indicated the sensitivity of the analytical method for detection of zinc, iron, copper, and manganese.

Robustness

Robustness of the Atomic Absorption Spectroscopy method was evaluated by introducing small deliberate changes in analytical parameters such as wavelength and instrumental conditions. Standard solutions of zinc, iron, copper, and manganese were analyzed under the modified conditions and the absorbance values were recorded. The percentage relative standard deviation (%RSD) was calculated and compared with the results obtained under normal conditions. The method was found to be robust as no significant variation in results was observed . **Organoleptic Evaluation:**

The prepared herbal face pack was evaluated for its physical characteristics like colour ,odour, texture, appearance and texture by visual inspection.

pH Determination:

About 1 g of the face pack powder was dissolved in 10 mL of distilled water and allowed to stand for some time. The pH of the solution was measured using a calibrated pH meter.

Particle Size Analysis:

The prepared powder was passed through standard sieve no. 80 to ensure uniform particle size distribution. Uniform particle size was obtained, which contributes to smooth texture and better application of the face pack.

Bulk Density:

A known quantity of powder was taken and poured into a graduated cylinder without tapping, and the volume occupied was recorded.

Tapped Density:

Tapped density of the prepared herbal face pack powder was determined to evaluate its packing ability and flow characteristics. A known quantity of the powder was accurately weighed and transferred into a clean, dry graduated measuring cylinder. The initial volume (bulk volume) was noted. The cylinder was then tapped repeatedly (usually 100–500 taps) using a tapped density apparatus or manually by gently tapping on a firm surface until a constant volume was obtained. The final tapped volume was recorded.

Tapped density was calculated using the formula: Tapped density=Mass of powder/Tapped volume

Moisture Content (Loss on Drying):

The moisture content of the prepared herbal face pack was determined by the Loss on Drying method. A known quantity of the formulation (about 1–2 g) was accurately weighed and placed in a clean, dry evaporating dish. The sample was then dried in a hot air oven at 105°C until a constant weight was obtained. After drying, the sample was cooled in a desiccator and reweighed. The percentage moisture content was calculated using the formula: Moisture Content (%) = Initial weight - Final weight / Initial weight x 100 10) Stability Studies: The prepared formulation was stored at room temperature and observed over a period for any changes in colour , odour, texture, or appearance

Results and Discussion

The analytical method was optimized to achieve maximum extraction efficiency and accurate quantification of iron, copper, manganese, and zinc from the developed formulations. Three formulations (F1, F2, and F3) were prepared and evaluated. Approximately 1 g of each formulation was dispersed in distilled water and subjected to sonication for 10–15 minutes to facilitate the release of metal ions into the extraction medium. The solutions were filtered through Whatman filter paper and diluted to 50 mL with distilled water. Various extraction conditions were evaluated, including sample amount, extraction volume, sonication time, and dilution factor. The optimized conditions consisted of 1 g sample weight, sonication for 15 minutes, filtration through Whatman filter paper, and dilution to 50 mL. These optimized conditions provided clear solutions with consistent absorbance values and satisfactory recovery of all analytes shown in **Table no 2.** Colour of the formulation is found to be yellowish to light brown ,Odour is pleasant ,Texture is Fine, smooth powder ,appearance is homogeneous and the texture was smooth ,ease to spread, were observed visually and by touch.

Table No 2: Optimized Conditions for AAS Method

Parameter	**Optimized Condition**
Instrument	Atomic Absorption Spectrophotometer (Thermo Fisher Scientific)
Type	Air–acetylene
Analytical Wavelength for Zn	213.9 nm
Analytical Wavelength for Cu	324.8nm

Analytical Wavelength for Mn	279.5nm
Analytical Wavelength for Fe	248.3nm
Working concentration range	2-10 µg/mL
Calibration Levels	2,4,6,8 and 10 µg/mL
Number of Calibration points	5
Replicates for Precision study	3
Sample Weight	1g
Extraction solvent	Distilled water
Sonication time	10-15 min
Filtration medium	Whatman filter paper no. 1
Final Volume	50ml
Sample Formulations	F1,F2, and F3
Analytes Determined	Fe, Cu, Mn and Zn
Aspiration Rate	5 ml/min
Slit Width	0.2-0.5 nm
Lamp Current	As recommended by manufacturer
Validation Parameters	Linearity, Accuracy, Precision, LOD<LOQ and Robustness
Acceptance Criteria for precision	% RSD ≤ 2%
Selected Optimized Method	Sonication-assisted aqueous extraction followed by AAS analysis

Concentration of Trace Elements in Different Herbal Face Pack Formulations

The concentrations of iron (Fe), zinc (Zn), manganese (Mn), and copper (Cu) in the herbal face pack formulations are presented in Table . Among the formulations, F2 showed the highest levels of all trace elements, with Fe (0.67 ppm), Zn (0.20 ppm), Mn (0.32 ppm), and Cu (0.62 ppm). F3 exhibited intermediate concentrations, while F1 contained the lowest levels of these elements.Iron was the most abundant trace element in all formulations, followed by copper, manganese, and zinc. The presence of these essential trace elements may contribute to skin nourishment and protection. Overall, F2 demonstrated a comparatively higher trace element content than F1 and F3.

Table No 3: Concentration of Trace Elements in Different Herbal Face Pack Formulations.

Formulation	Fe(ppm)	Zn(ppm)	Mn(ppm)	Cu(ppm)
F1	0.55	0.15	0.25	0.58
F2	0.67	0.20	0.32	0.62
F3	0.60	0.18	0.28	0.60

Linearity

The linearity of the analytical method was evaluated by plotting calibration curves of absorbance versus concentration for iron (Fe), manganese (Mn), zinc (Zn), and copper (Cu). The calibration curves exhibited a direct proportional relationship between concentration and absorbance within the studied concentration range, indicating good linearity of the method. Figures . show the linearity curves for Fe, Mn, Zn, and Cu, respectively. The obtained results demonstrate that the analytical method is suitable for the accurate quantification of these trace elements in herbal face pack formulations.

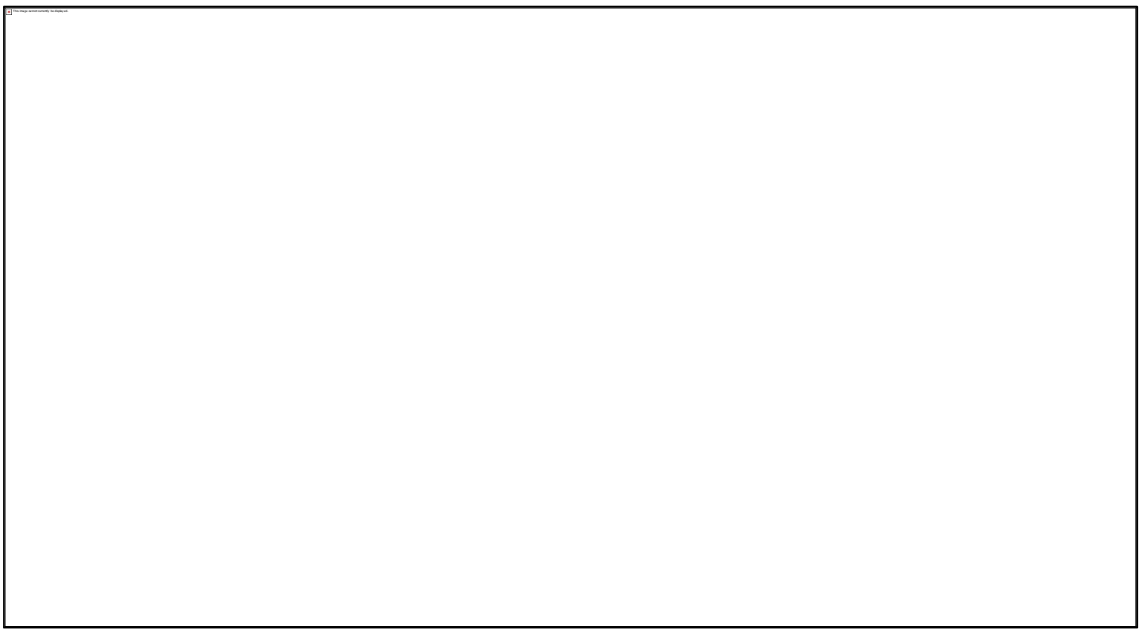

Figure 1. Linearity plot of Fe (A), Zn (B), Cu (C), Mn (D)

Accuracy

Recovery studies were performed at 80%, 100%, and 120% levels for Fe, Zn, Cu, and Mn. The percentage recoveries ranged from 94.50–98.17% for Fe, 90.75–100.20% for Zn, 93.00–96.70% for Cu, and 94.00–100.75% for Mn. The recovery results were within acceptable limits, confirming the accuracy of the method for trace element analysis.

Table No 4: Accuracy

Level	Amount Added (ppm)	Amount Found (ppm)	Recovery(%)
Fe			
80%	8	7.56	94.50
100%	10	9.64	96.40
120%	12	11.78	98.17
Zn			
80%	8	7.26	90.75
100%	10	10.02	100.20
120%	12	11.72	97.67
Cu			
80%	8	7.44	93.0
100%	10	9.67	96.70
120%	12	11.44	95.33
Mn			
80%	8	8.06	100.75
100%	10	9.40	94.0
120%	12	11.48	95.67

Precision

The precision of the developed AAS method was evaluated by performing intraday and interday precision studies at five concentration levels (2, 4, 6, 8, and 10 µg/mL) for Zn, Cu, Mn, and Fe. Each concentration was analyzed in triplicate, and the results were expressed as mean absorbance, standard deviation (SD), and percentage relative standard deviation (%RSD). The %RSD values for all elements were found to be less than 2%, indicating good precision and repeatability of the developed method. The results demonstrate that the method is precise and suitable for the quantitative determination of Zn, Cu, Mn, and Fe in the polyherbal face pack formulation.

Intraday Precision Study

Table No 5: Intraday Precision Study for Zn and Cu

Concentration (µg/mL)	Zn			Cu		
	Absorbance	Mean± SD	%RSD	Absorbance	Mean± SD	%RSD
2	0.125	0.125±0.002	1.60	0.103	0.103±0.001	1.15
	0.127			0.104		
	0.124			0.102		
4	0.248	0.249±0.002	0.80	0.202	0.202±0.002	0.99
	0.251			0.204		
	0.249			0.201		
6	0.373	0.374±0.002	0.53	0.301	0.302±0.002	0.66
	0.376			0.304		
	0.372			0.300		
8	0.498	0.499±0.003	0.60	0.403	0.404±0.002	0.54
	0.502			0.407		
	0.497			0.402		
10	0.622			0.502		
	0.626	0.623±0.003	0.48	0.505	0.502±0.003	0.60
	0.621			0.499		

Table No 6: Intraday Precision Study for Mn and Fe

Concentration (µg/mL)	Mn			Fe		
	Absorbance	Mean± SD	%RSD	Absorbance	Mean± SD	%RSD
2	0.078	0.078 ±0.01	1.28	0.192	0.192±0.002	1.04
	0.079			0.194		

	0.077			0.191		
4	0.156	0.156 ±0.002	1.08	0.383	0.384±0.002	0.52
	0.158			0.386		
	0.155			0.382		
6	0.234	0.234 ±0.002	0.80	0.573	0.574±0.002	0.45
	0.236			0.576		
	0.232			0.572		
8	0.312	0.312 ±0.003	0.96	0.764	0.765±0.004	0.49
	0.315			0.769		
	0.310			0.761		
10	0.388			0.954		
	0.392	0.389 ±0.003	0.77	0.961	0.956±0.005	0.52
	0.387			0.952		

Interday Precision Study:

Table No 7: Interday Precision Study for Zn and Cu

Concentration	**Zn**			**Cu**		
	Absorbance	Mean± SD	%RSD	Absorbance	Mean± SD	%RSD
2	0.125	0.125±0.002	1.60	0.103	0.103±0.001	1.15
	0.127			0.104		
	0.124			0.102		
4	0.248	0.249±0.002	0.80	0.202	0.202±0.002	0.99
	0.251			0.204		
	0.249			0.201		
6	0.373	0.37±0.002	0.53	0.301	0.302±0.02	0.66
	0.376			0.304		
	0.372			0.300		

8	0.498	0.499±0.003	0.60	0.403	0.404±0.002	0.54
	0.502			0.407		
	0.497			0.402		
10	0.622	0.623±0.003	0.48	0.502	0.502±0.003	0.60
	0.626			0.505		
	0.621			0.499		

Table No 8: Interday Precision Study for Mn and Fe

Concentration	**Mn**			**Fe**		
	Absorbance	Mean± SD	%RSD	Absorbance	Mean± SD	%RSD
2	0.078	0.078±0.001	1.28	0.192	0.192±0.002	1.04
	0.079			0.194		
	0.077			0.191		
4	0.156	0.156±0.02	1.08	0.383	0.384±0.002	0.52
	0.158			0.386		
	0.155			0.382		
6	0.234	0.234±0.002	0.80	0.573	0.574±0.002	0.45
	0.236			0.576		
	0.232			0.572		
8	0.312	0.312±0.003	0.96	0.764	0.765±0.004	0.49
	0.315			0.769		
	0.310			0.761		
10	0.388	0.389±0.003	0.77	0.954	0.956±0.005	0.52
	0.392			0.961		
	0.387			0.952		

Robustness

The robustness of the developed AAS method was evaluated by introducing small deliberate variations in analytical parameters, including wavelength (±0.2–0.3 nm) and flow rate (±0.1 L/min) for the determination of Zn, Fe, Cu, and Mn. The absorbance values obtained under the modified conditions showed minimal variation, with %RSD values ranging from 0.48% to 1.05%. Since all %RSD values were below 2%, the method was found to be robust and unaffected by minor changes in experimental conditions. These results demonstrate the reliability of the developed method for routine elemental analysis.

ROBUSTNESS:

Zinc (Zn)

Table No 9: Robustness Study for Zinc (Zn)

Parameter	Change	Absorbance	Mean ± SD	%RSD
Wavelength	213.7 nm	0.412	0.414 ± 0.002	0.48
Wavelength	213.9 nm	0.416	0.414 ± 0.002	0.48
Wavelength	214.1 nm	0.414	0.414 ± 0.002	0.48
flow rate	1.4 L/min	0.409	0.411 ± 0.003	0.73
flow rate	1.5 L/min	0.412	0.411 ± 0.003	0.73
flow rate	1.6 L/min	0.412	0.411 ± 0.003	0.73

Iron (Fe)

Table No 10: Robustness Study for Iron (Fe)

Parameter	Change	Absorbance	Mean ± SD	%RSD
Wavelength	248.0 nm	0.536	0.538 ± 0.003	0.56
Wavelength	248.3 nm	0.540	0.538 ± 0.003	0.56
Wavelength	248.6 nm	0.538	0.538 ± 0.003	0.56
flow rate	1.4 L/min	0.531	0.533 ± 0.003	0.56
flow rate	1.5 L/min	0.535	0.533 ± 0.003	0.56
flow rate	1.6 L/min	0.533	0.533 ± 0.003	0.56

Copper (Cu)

Table No 11: Robustness Study for Copper (Cu)

Parameter	Change	Absorbance	Mean ± SD	%RSD
Wavelength	324.6 nm	0.286	0.287 ± 0.002	0.70
Wavelength	324.8 nm	0.289	0.287 ± 0.002	0.70
Wavelength	325.0 nm	0.286	0.287 ± 0.002	0.70
flow rate	1.4 L/min	0.282	0.284 ± 0.003	1.05
flow rate	1.5 L/min	0.285	0.284 ± 0.003	1.05

flow rate	1.6 L/min	0.285	0.284 ± 0.003	1.05

Manganese (Mn)

Table No 12: Robustness Study for Manganese (Mn)

Parameter	Change	Absorbance	Mean ± SD	%RSD
Wavelength	279.2 nm	0.198	0.199 ± 0.002	1.00
Wavelength	279.5 nm	0.201	0.199 ± 0.002	1.00
Wavelength	279.8 nm	0.198	0.199 ± 0.002	1.00
flow rate	1.4 L/min	0.195	0.197 ± 0.002	1.01
flow rate	1.5 L/min	0.199	0.197 ± 0.002	1.01
flow rate	1.6 L/min	0.197	0.197 ± 0.002	1.01

Specificity

The specificity of the developed AAS method was evaluated for the determination of Zn, Fe, Cu, and Mn in the polyherbal face pack formulation. The method showed no interference from other formulation components. The results obtained were consistent with low standard deviation values, confirming that the method is specific and suitable for accurate elemental analysis.

Table No 13: Specificity Results for Zn, Fe, Cu and Mn

Sample	Absorbance	Concentration (µg/mL)	% Element	Mean ± S.D
Zinc	0.412	4.12	0.0412	0.041 ± 0.002
	0.418	4.18	0.0418	0.041 ± 0.002
	0.409	4.09	0.0409	0.041 ± 0.002
Iron	0.536	5.36	0.0536	0.054 ± 0.003
	0.542	5.42	0.0542	0.054 ± 0.003
	0.531	5.31	0.0531	0.054 ± 0.003
Copper	0.286	2.86	0.0286	0.029 ± 0.001
	0.291	2.91	0.0291	0.029 ± 0.001
	0.284	2.84	0.0284	0.029 ± 0.001
Manganese	0.198	1.98	0.0198	0.020 ± 0.001
	0.202	2.02	0.0202	0.020 ± 0.001
	0.196	1.96	0.0196	0.020 ± 0.001

LOD and LOQ

The LOD and LOQ of the developed AAS method were determined for Zn, Cu, Mn, and Fe. The low LOD and LOQ values obtained indicate that the method is sufficiently sensitive for the detection and quantification of trace elements in the polyherbal face pack formulation.

Table No 14: LOD and LOQ Values for Zn, Fe, Cu and Mn for facepack

Set	Zn Zn Slope		Cu Cu Slope		Mg Mg Slope		Fe Fe Slope	
	R^2	Absorban Ce	R^2	Absorban ce	R^2	Absorban ce	R^2	Absorban ce
1	0.9991	0.0521	0.9988	0.0415	0.9990	0.0382	0.9992	0.0584

2	0.9990	0.0523	0.9989	0.0413	0.9988	0.0385	0.9991	0.0587
3	0.9992	0.0520	0.9990	0.0416	0.9991	0.0383	0.9993	0.0585
4	0.9989	0.0522	0.9987	0.0414	0.9989	0.0384	0.9990	0.0586
5	0.9991	0.0521	0.9988	0.0415	0.9990	0.0382	0.9992	0.0585
Mean	0.9990	0.0521	0.9988	0.0414	0.9989	0.0383	0.9991	0.0585
SD	0.0001	0.0001	0.0001	0.0001	0.0001	0.0001	0.0001	0.0001
LOD	0.012		0.015		0.011		0.010	
LOQ	0.038		0.048		0.035		0.033	

4. CONCLUSION

The present study successfully developed and evaluated a polyherbal face pack containing multani mitti, red lentil, gram flour, lodhra, turmeric, orange peel, and aloe vera. Among the three formulations prepared, formulation F2 was found to be the most suitable based on its physicochemical characteristics, skin-compatible pH, and absence of any skin irritation. A simple, accurate, precise, specific, robust, and sensitive Atomic Absorption Spectroscopy (AAS) method was successfully developed and validated for the determination of zinc, iron, copper, and manganese in the formulation. The validated method demonstrated excellent linearity, recovery, precision (%RSD < 2%), and satisfactory LOD and LOQ values. Elemental analysis revealed that all trace element concentrations were within acceptable safety limits, confirming the non-toxic nature of the formulation. Therefore, the developed polyherbal face pack can be considered a safe, effective, economical, and scientifically standardized herbal cosmetic product for skincare applications. The validated AAS method can also serve as a reliable tool for routine quality control and elemental standardization of herbal cosmetic formulations.

Author contribution

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Conflict of Interest

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