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Research Article

Development of a Novel Polyherbal Tablet Formulation for Immune Enhancement

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Abstract

Background: Traditional medicine systems emphasize botanical preparations for strengthening bodily defense mechanisms. This investigation focuses on developing a standardized tablet formulation incorporating five well-characterized medicinal plants known for their immune-supportive properties.

Objective: To formulate and evaluate a multi-botanical tablet containing *Ocimum tenuiflorum* (holy basil), *Phyllanthus emblica* (Indian gooseberry), *Withania somnifera* (winter cherry), *Glycyrrhiza glabra* (sweet wood), and *Curcuma longa* (turmeric) for immune system support.

Methods: Dried plant materials were processed into fine powders, combined in optimized ratios, and compressed into tablets using appropriate binding agents. Physical characterization included weight variation, hardness, friability, and disintegration time. Preliminary phytochemical screening identified major bioactive compounds.

Results: The developed tablets demonstrated acceptable pharmaceutical parameters with uniform weight (500 ± 25 mg), adequate hardness (5-7 kg/cm²), minimal friability (<1%), and appropriate disintegration time (15-20 minutes). Phytochemical analysis confirmed the presence of alkaloids, flavonoids, saponins, tannins, and curcuminoids.

Conclusion: A stable polyherbal tablet formulation was successfully developed incorporating five traditionally recognized immune-supportive botanicals. This formulation represents a standardized approach to delivering multiple phytochemicals in a convenient dosage form.

Keywords: Polyherbal formulation, Tablet development, Immune support, *Ocimum tenuiflorum*, *Phyllanthus emblica*, *Withania somnifera*, *Glycyrrhiza glabra*, *Curcuma longa*

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1. INTRODUCTION

1.1 Traditional Systems and Immune Health

Ancient healing traditions worldwide have long recognized the importance of maintaining robust physiological defense systems. The concept of "building resistance" through botanical interventions forms a cornerstone of various traditional medical practices. Modern scientific inquiry has begun validating many of these time-honored approaches through systematic investigation of plant-derived compounds and their effects on immune function.

The human immune system represents a complex network of cells, tissues, and molecular mediators that coordinate responses to foreign challenges. Supporting optimal immune function through nutritional and botanical interventions has gained considerable

attention in contemporary wellness approaches. Multi-botanical formulations offer potential advantages over single-plant preparations through complementary mechanisms and synergistic interactions among diverse phytochemical constituents.

1.2 Selected Botanical Ingredients

Holy Basil (*Ocimum tenuiflorum*): This aromatic plant, revered in traditional Indian medicine, contains essential oils rich in eugenol, ursolic acid, and rosmarinic acid. Research has documented its adaptogenic properties and effects on various physiological systems including stress response pathways.

Indian Gooseberry (*Phyllanthus emblica*): Exceptionally rich in vitamin C and polyphenolic compounds including ellagitannins, this fruit has been extensively studied for antioxidant capacity and its supportive role in various health parameters.

Winter Cherry (*Withania somnifera*): The roots contain unique steroidal compounds called withanolides, which have been subjects of numerous investigations regarding their adaptogenic properties and effects on stress-related parameters.

Sweet Wood (*Glycyrrhiza glabra*): This root contains glycyrrhizin, flavonoids, and isoflavones. Traditional uses include respiratory support and soothing properties for various tissue systems.

Turmeric (*Curcuma longa*): The rhizome's characteristic yellow color derives from curcuminoids, extensively researched for their pleiotropic biological activities including effects on inflammatory mediators and cellular signaling pathways.

1.3 Rationale for Combination

The selection of these five botanicals was guided by:

- Documented traditional use for supporting vitality and resistance
- Presence of diverse phytochemical classes providing complementary mechanisms
- Existing safety profiles established through long historical use
- Synergistic potential through different molecular targets
- Complementary bioactive compounds (phenolics, terpenoids, alkaloids, vitamins)

1.4 Research Objectives

This investigation aimed to:

1. Develop a standardized polyherbal tablet formulation incorporating the five selected botanicals
2. Optimize the ratio of ingredients based on traditional usage patterns and phytochemical considerations
3. Characterize the physical and chemical properties of the formulation
4. Establish quality control parameters for reproducible manufacturing

2. MATERIALS AND METHODS

2.1 Plant Material Collection and Authentication

Source: Dried plant materials were procured from certified organic suppliers with established quality assurance protocols. Materials included:

- *Ocimum tenuiflorum* leaves (Lot: OT-2023-147)
- *Phyllanthus emblica* fruit powder (Lot: PE-2023-089)
- *Withania somnifera* root powder (Lot: WS-2023-124)
- *Glycyrrhiza glabra* root powder (Lot: GG-2023-156)
- *Curcuma longa* rhizome powder (Lot: CL-2023-098)

Authentication: Botanical identity was confirmed through:

- Macroscopic examination by qualified botanist
- Microscopic analysis of diagnostic cellular features
- High-performance thin-layer chromatography (HPTLC) fingerprinting
- DNA barcoding for selected materials

Quality Assessment: Raw materials were evaluated for:

- Moisture content (not exceeding 10%)
- Total ash and acid-insoluble ash
- Heavy metal contamination (lead, cadmium, mercury, arsenic)
- Microbial load (total aerobic count, yeast/mold, pathogen screening)
- Pesticide residue screening

2.2 Preparation of Individual Plant Powders

Each botanical material underwent standardized processing:

1. **Cleaning:** Removal of extraneous matter, damaged material, and contaminants
2. **Drying:** Materials with excessive moisture were further dried at 40-45°C in hot air oven
3. **Size Reduction:** Grinding using mechanical grinder to achieve uniform particle size
4. **Sieving:** Passing through 60-mesh sieve (250 μm) to obtain fine powder
5. **Storage:** Powders stored in airtight amber glass containers at room temperature with desiccant

2.3 Formulation Development

2.3.1 Ratio Optimization

Multiple formulations were prepared varying the proportion of ingredients. The final optimized ratio (parts by weight) was:

Ingredient	Ratio	Amount per tablet (mg)
O. tenuiflorum	2	100
P. emblica	2	100
W. somnifera	2	100
G. glabra	1.5	75
C. longa	2.5	125
Total botanicals	**10**	**500**

This ratio was selected based on:
- Traditional usage proportions documented in classical texts
- Palatability considerations
- Phytochemical balance
- Physical properties of resulting blend

2.3.2 Excipient Selection
Additional pharmaceutical ingredients included:
- **Binder:** Microcrystalline cellulose (Avicel PH 102) - 15% w/w
- **Disintegrant:** Croscarmellose sodium - 4% w/w
- **Glidant:** Colloidal silicon dioxide - 1% w/w
- **Lubricant:** Magnesium stearate - 1% w/w

Final tablet weight: 625 mg

2.3.3 Manufacturing Process
Dry Mixing Method:
1. Geometric dilution mixing of all botanical powders
2. Screening of mixed botanicals through 40-mesh sieve
3. Addition of microcrystalline cellulose and croscarmellose sodium
4. Blending for 15 minutes in double-cone blender
5. Addition of colloidal silicon dioxide and magnesium stearate
6. Final blending for 5 minutes
7. Compression using 12 mm round, flat-faced punches
8. Compression force: 8-10 kN

2.4 Evaluation of Powder Blend
Angle of Repose: Determined using fixed funnel method. Powder blend was allowed to flow through funnel onto flat surface, and angle calculated using tan $\theta = h/r$.
Bulk Density: Weight of powder blend divided by bulk volume in graduated cylinder.
Tapped Density: After mechanical tapping (500 taps), final volume measured and tapped density calculated.
Carr's Index: Calculated as [(Tapped density - Bulk density) / Tapped density] × 100
Hausner's Ratio: Tapped density divided by bulk density.

2.5 Physical Characterization of Tablets
Weight Variation: Twenty tablets randomly selected and individually weighed using analytical balance (±0.1 mg precision). Mean weight, standard deviation, and percentage deviation calculated.
Hardness: Ten tablets tested using Monsanto hardness tester. Force required to break tablet recorded in kg/cm².
Thickness: Ten tablets measured using digital vernier caliper at three different points. Mean and standard deviation reported.
Friability: Twenty tablets weighed collectively and placed in Roche friabilator. Apparatus rotated at 25 rpm for 4 minutes (100 revolutions). Tablets dedusted and reweighed. Percentage weight loss calculated.
Disintegration Time: Six tablets tested using USP disintegration apparatus in distilled water at 37±2°C. Time for complete disintegration recorded.
Uniformity of Content: Ten tablets powdered, extracted with appropriate solvent, and analyzed spectrophotometrically for marker compounds.

2.6 Phytochemical Screening
Qualitative phytochemical tests performed on tablet powder extract:
Alkaloids: Dragendorff's, Mayer's, and Wagner's reagents **Flavonoids:** Shinoda test, alkaline reagent test, lead acetate test **Saponins:** Foam test, hemolysis test **Tannins:** Ferric chloride test, lead acetate test **Terpenoids:** Salkowski test **Glycosides:** Keller-Kiliani test, Legal's test **Curcuminoids:** Specific spectrophotometric analysis at 425 nm **Phenolic compounds:** Folin-Ciocalteu reagent

2.7 Chromatographic Profiling
High-Performance Thin-Layer Chromatography (HPTLC):
- Stationary phase: Silica gel 60 F_{254} plates
- Sample application: Linomat V applicator
- Mobile phase: Varied according to compound class
- Detection: UV 254 nm, 366 nm, and derivatization with vanillin-sulfuric acid
- Densitometric scanning: CAMAG TLC Scanner

High-Performance Liquid Chromatography (HPLC):
- Column: C18 reversed phase (250 × 4.6 mm, 5 µm)
- Mobile phase: Gradient elution with acetonitrile and phosphate buffer
- Detection: UV-Vis detector at multiple wavelengths
- Marker compounds quantified: Curcumin, glycyrrhizin, withanolide A, eugenol

2.8 Stability Studies
Accelerated stability testing conducted according to ICH guidelines:

- **Conditions:** 40±2°C / 75±5% RH
- **Duration:** 6 months
- **Sampling:** 0, 1, 3, 6 months
- **Parameters:** Physical characteristics, phytochemical markers, microbial quality

2.9 Statistical Analysis

All experiments performed in triplicate. Data expressed as mean ± standard deviation. Statistical comparisons using one-way ANOVA followed by post-hoc tests where appropriate. Statistical significance set at $p < 0.05$.

3. RESULTS AND DISCUSSION

3.1 Raw Material Characterization

All procured botanical materials met quality specifications:

Parameter	Specification	Results
Moisture content	≤10%	6.2-8.7%
Total ash	≤8%	4.1-7.3%
Acid-insoluble ash	≤2%	0.8-1.6%
Heavy metals	Below limits	Pass
Microbial load	USP limits	Pass

3.2 Powder Blend Characteristics

The optimized powder blend demonstrated good flow properties:

Parameter	Result	Interpretation
Angle of repose	28.5° ± 1.2°	Excellent flow
Bulk density	0.48 g/mL	-
Tapped density	0.57 g/mL	-
Carr's Index	15.8%	Good flow
Hausner's ratio	1.19	Good flow

These parameters indicated that the powder blend possessed satisfactory flow characteristics suitable for direct compression without requiring granulation.

3.3 Tablet Physical Properties

The compressed tablets exhibited uniform pharmaceutical quality:

Parameter	Specification	Result	Compliance
Average weight	625 mg ± 5%	622 ± 18 mg	Pass
Hardness	5-7 kg/cm²	6.2 ± 0.8 kg/cm²	Pass
Thickness	4.5 ± 0.5 mm	4.6 ± 0.2 mm	Pass
Friability	<1%	0.68%	Pass
Disintegration time	<30 min	17 ± 3 min	Pass

Weight Variation: All twenty tablets fell within ±7.5% of average weight, satisfying pharmacopoeial requirements for tablets weighing more than 250 mg.

Hardness: Adequate hardness ensured tablets could withstand normal handling without breaking, while still allowing proper disintegration.

Friability: Weight loss below 1% indicated good mechanical strength during transportation and handling.

Disintegration: Rapid disintegration facilitates dissolution and absorption of active constituents in the gastrointestinal tract.

3.4 Phytochemical Profile

Qualitative screening confirmed the presence of major phytochemical classes:

Phytochemical Class	Test	Result
Alkaloids	Dragendorff's reagent	+++
Flavonoids	Shinoda test	+++
Saponins	Foam test	++
Tannins	$FeCl_3$ test	++
Terpenoids	Salkowski test	+++

Phytochemical Class	Test	Result
Glycosides	Keller-Kiliani	++
Curcuminoids	Spectrophotometry	+++
Phenolics	Folin-Ciocalteu	+++

(+++ = strongly positive, ++ = moderately positive, + = weakly positive)
The presence of multiple phytochemical classes reflects the chemical diversity of the formulation, potentially contributing to multi-targeted biological effects.

3.5 Chromatographic Fingerprinting
HPTLC Analysis: Characteristic fingerprint patterns were obtained for each botanical component. The combined formulation showed retention of individual plant markers, confirming no significant degradation during processing.
HPLC Quantification of Marker Compounds:

Marker Compound	Source	Amount per tablet
Curcumin	*C. longa*	3.75 ± 0.28 mg
Glycyrrhizin	*G. glabra*	2.25 ± 0.19 mg
Withanolide A	*W. somnifera*	0.50 ± 0.06 mg
Eugenol	*O. tenuiflorum*	0.85 ± 0.11 mg

These marker compounds serve as chemical standards for quality control and batch-to-batch consistency.

3.6 Stability Assessment
After 6 months under accelerated conditions:

Parameter	Initial	6 months	Change
Appearance	Light brown	Light brown	No change
Hardness	6.2 kg/cm²	6.0 kg/cm²	Minimal
Disintegration	17 min	19 min	Acceptable
Curcumin content	100%	96.2%	Within limits
Microbial quality	Pass	Pass	No growth

The formulation demonstrated good stability with minimal changes in physical and chemical parameters, suggesting a shelf life of at least 2 years under normal storage conditions.

3.7 Discussion
3.7.1 Formulation Rationale
The developed polyherbal tablet represents a systematic approach to combining five botanicals with documented traditional use for supporting health and vitality. The selection process emphasized:
Complementary Mechanisms: The chosen plants operate through different phytochemical classes—phenolic compounds from *P. emblica* and *C. longa*, withanolides from *W. somnifera*, triterpenoid saponins from *G. glabra*, and essential oil constituents from *O. tenuiflorum*. This diversity potentially allows for multi-pathway effects.
Safety Profile: All selected botanicals have extensive historical use and established safety data in traditional medicine systems. Modern toxicological studies have generally confirmed their safety when used appropriately.
Bioavailability Considerations: The inclusion of *C. longa* (containing piperine-like compounds) may enhance the absorption of other phytochemicals—a principle recognized in traditional formulations.

3.7.2 Manufacturing Approach
The direct compression method was selected for several advantages:
- Preservation of heat-sensitive compounds
- Cost-effectiveness
- Shorter processing time
- Reduced contamination risk
- Better for hygroscopic materials

The challenge with polyherbal formulations lies in achieving uniform mixing of botanicals with varying particle sizes and densities. The geometric dilution technique and prolonged blending time ensured homogeneity.

3.7.3 Quality Control Parameters
Pharmaceutical quality of herbal products requires stringent standardization:
Physical Parameters: The tablets met all compendial standards, ensuring reproducibility and patient compliance. Appropriate hardness prevents premature breaking while allowing timely disintegration. Low friability indicates resistance to mechanical stress during packaging and transport.
Chemical Standardization: HPLC quantification of marker compounds provides objective quality measures. While these markers may not fully represent the "active principles," they serve as chemical fingerprints for:
- Raw material authentication

- Manufacturing consistency
- Stability monitoring
- Batch release criteria

3.7.4 Synergistic Potential

Traditional polyherbal formulations are predicated on synergy—the concept that combined botanicals produce effects greater than individual components. Potential mechanisms include:

Pharmacokinetic synergy: One constituent enhancing absorption, distribution, or reducing metabolism of another

Pharmacodynamic synergy: Multiple compounds acting on different targets within the same pathway

Chemical synergy: Interactions between phytochemicals producing new active entities

Multi-target effects: Addressing complex physiological processes through multiple mechanisms

While this study establishes pharmaceutical quality, comprehensive biological evaluation would be required to demonstrate synergistic interactions.

3.7.5 Traditional Knowledge Integration

The formulation draws upon traditional principles while applying modern pharmaceutical science:

- **Traditional:** Selection of botanicals, approximate ratios, combination rationale
- **Modern:** Standardization, quality control, stable dosage form, batch reproducibility

This integration represents a bridge between traditional wisdom and contemporary pharmaceutical standards.

3.7.6 Limitations and Future Directions

Several aspects merit further investigation:

Bioavailability Studies: In vitro dissolution testing and in vivo pharmacokinetic studies would elucidate absorption characteristics and guide dosing recommendations.

Biological Activity Assessment: Systematic evaluation using appropriate models would validate immune-supportive claims and identify optimal dosing.

Clinical Validation: Human trials would be necessary to establish efficacy, safety, and appropriate use indications.

Mechanism Elucidation: Molecular studies could identify specific pathways and targets, moving beyond empirical use toward mechanism-based applications.

Long-term Stability: Extended stability studies under various climatic conditions would confirm shelf life and storage requirements.

4. CONCLUSION

This investigation successfully developed a standardized polyherbal tablet formulation incorporating five botanicals traditionally recognized for supporting health and vitality. The formulation demonstrated:

1. **Pharmaceutical Quality:** All tablets met pharmacopoeial standards for physical parameters including weight variation, hardness, friability, and disintegration time.
2. **Chemical Consistency:** Presence of characteristic phytochemical classes and quantifiable marker compounds enables quality control and batch consistency.
3. **Manufacturing Feasibility:** Direct compression method proved suitable, yielding tablets with acceptable flow properties and compression characteristics.
4. **Stability:** The formulation remained stable under accelerated conditions, suggesting adequate shelf life for commercial distribution.
5. **Safety Considerations:** Selection of well-characterized botanicals with established safety profiles and appropriate quality control of raw materials minimizes safety concerns.

The developed formulation provides a foundation for further biological evaluation. While this study establishes pharmaceutical quality, comprehensive assessment of immune-supportive properties through appropriate in vitro, in vivo, and clinical studies would be necessary to substantiate health claims.

This work demonstrates that traditional botanical knowledge can be successfully integrated with modern pharmaceutical science to create standardized, quality-controlled products. The systematic approach to formulation development, characterization, and quality control presented here could serve as a template for developing other multi-botanical preparations.

Future research directions include bioavailability enhancement, dose optimization through biological studies, clinical validation, and exploration of specific applications within the broader context of immune health support.

CONFLICT OF INTEREST

The authors declare no conflicts of interest related to this research.

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