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Review Article

Clitoria ternatea L.: Pharmaceutical Quality Assessment, Ternatin Standardization, Extraction Technologies and Formulation Stability

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ABSTRACT

Clitoria ternatea L. (Fabaceae), commonly known as butterfly pea or aparajita, is a medicinal and food-use plant whose blue flowers are a notable source of polyacylated anthocyanins known as ternatins. Interest in the plant has expanded from traditional use and preliminary pharmacological screening to natural colourant development, botanical preparations and pharmaceutical research. However, the quality of C. ternatea preparations can vary with botanical identity, plant part, cultivar, harvest conditions, drying, extraction solvent and processing conditions. This review focuses on pharmaceutical quality assessment rather than repeating a general catalogue of therapeutic claims. It discusses botanical authentication, major phytochemical markers, conventional and advanced extraction approaches, chromatographic and spectroscopic analysis, standardization parameters, pigment stability, formulation considerations and safety-related quality requirements. Water and aqueous ethanol are relevant solvents for polar flower pigments, while extraction conditions must be optimized to limit degradation. TLC and HPTLC can support fingerprinting, whereas validated HPLC and LC–MS methods are more suitable for marker profiling and characterization. UV–visible spectroscopy can monitor pigment behaviour but should not be used alone to establish chemical identity. Existing literature supports the potential of C. ternatea as a source of natural pigments and bioactive constituents; nevertheless, standardized reference materials, validated analytical methods, batch-to-batch studies and product-specific safety data remain important needs. The review identifies practical quality-control priorities for the development of reproducible flower extracts and finished formulations.

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INTRODUCTION

Clitoria ternatea L. is a perennial climbing legume belonging to the family Fabaceae. It is commonly known as butterfly pea or aparajita and is cultivated in tropical and subtropical regions. The plant has attracted attention because of its traditional use, distinctive blue or white flowers and diverse chemical constituents. The flowers contain water-soluble, polyacylated anthocyanins known as ternatins, which contribute to their characteristic blue colour and pH-dependent colour behaviour.

Published reviews have discussed the ethnomedicinal uses, phytochemistry and pharmacological activities of the species. More recent work has also examined its industrial and food applications. In this context, a pharmaceutical-quality perspective is useful because a botanical extract cannot be considered reproducible unless its identity, plant part, preparation method, chemical profile and quality attributes are clearly defined.25 Nov 2026

The composition of C. ternatea is not uniform across the whole plant. Flowers, leaves, roots and seeds may contain different metabolite profiles. Therefore, findings from one plant part should not automatically be applied to another. This review emphasizes flower-extract quality while referring to other plant parts only where relevant to authentication or analytical control.

2. REVIEW SCOPE AND LITERATURE APPROACH

This narrative review draws on published primary studies, established reviews and recognized guidance relevant to C. ternatea, anthocyanin extraction, herbal-material quality and analytical standardization. The topics considered include botanical authentication, phytochemical markers, extraction technologies, analytical methods, stability, formulation development and safety-related quality control.

The discussion distinguishes established analytical or compositional observations from proposed development approaches. It does not claim that a new experimental extraction or formulation has been performed. No quantitative meta-analysis was attempted because extraction procedures, plant materials and reported outcomes vary substantially between studies.

3. BOTANICAL IDENTITY AND RAW-MATERIAL AUTHENTICATION

The accepted botanical name is Clitoria ternatea L., family Fabaceae. The plant is a twining or climbing herb with compound leaves, papilionaceous flowers and flattened pods containing seeds. Flower colour may vary, and blue and white forms are reported. Correct identification is the first requirement for quality assessment because substitution, admixture or incorrect common-name use can affect the chemical profile of the final extract.

3.1 Taxonomic classification

Taxonomic rank	Classification
Kingdom	Plantae
Order	Fabales
Family	Fabaceae
Genus	Clitoria
Species	Clitoria ternatea L.

3.2 Collection and authentication

A raw-material record should include the accepted botanical name, family, plant part, flower colour or cultivar when known, collection location, collection date, season, collector and method of authentication. A voucher specimen should be deposited in a recognized herbarium where possible. Material should be collected from a traceable source and transported under conditions that minimize moisture accumulation and contamination.

3.3 Pharmacognostic quality parameters

Macroscopic and microscopic examination can help confirm identity and detect foreign matter. Additional quality parameters may include moisture content, total ash, acid-insoluble ash, water-soluble ash and extractive values, where appropriate to the monograph or study design. These tests should be selected and reported using a defined method; numerical limits should not be invented where an official specification is unavailable.

4. PHYTOCHEMICAL CONSTITUENTS AND QUALITY MARKERS

The chemical composition of C. ternatea depends on plant part, genotype or flower colour, geographical

origin, maturity, drying and extraction conditions. The best-known flower constituents are ternatins, a group of polyacylated delphinidin-derived anthocyanins. Flavonol glycosides and phenolic acids have also been reported. Other studies describe triterpenoids, sterols and cyclotides in different tissues.

4.1 Ternatins and anthocyanins

Anthocyanins are water-soluble pigments whose visible colour is influenced by pH and the surrounding chemical environment. Ternatins are structurally distinctive acylated anthocyanins associated with the intense blue colour of butterfly pea flowers. Their acylation and glycosylation patterns influence colour properties and stability. Because different ternatin species may occur together, total anthocyanin measurement alone does not provide a complete chemical fingerprint.

4.2 Flavonoids and phenolic compounds

Flavonol glycosides and phenolic acids contribute to the broader phytochemical profile. Compounds related to kaempferol and quercetin, together with phenolic-acid derivatives, have been discussed in the literature. Total phenolic or total flavonoid assays can provide supportive screening information, but they are non-specific and should be complemented by chromatographic analysis.

4.3 Cyclotides, triterpenoids and sterols

Cyclotides are small cyclic peptides characterized by a cyclic backbone and a cystine-knot arrangement of disulfide bonds. They are relevant to peptide and biotechnology research but should not be treated as equivalent to flower anthocyanins or assumed to be present at a defined level in every preparation. Triterpenoids and sterols are also reported in selected plant parts and may require different extraction and analytical procedures.

Constituent class	Representative group	Quality-assessment relevance
Anthocyanins	Ternatins / delphinidin-derived pigments	Key flower pigment markers; profile and stability assessment
Flavonols	Kaempferol- and quercetin-related glycosides	Supporting chromatographic fingerprint
Phenolic acids	Gallic, caffeic and p-coumaric acid derivatives	Supporting chemical characterization
Cyclotides	Cyclic cystine-knot peptides	Specialized peptide analysis; tissue-specific
Triterpenoids / sterols	Reported non-polar constituents	Require suitable solvent and separate marker strategy

5. EXTRACTION TECHNOLOGIES

Extraction conditions determine the yield, composition and stability of the final material. The solvent should be selected according to the intended marker compounds. Water and aqueous ethanol are commonly relevant for polar anthocyanins and other hydrophilic constituents. Solvent concentration, solid-to-liquid ratio, pH, temperature, extraction time, particle size, light exposure and oxygen contact should be documented.

5.1 Conventional extraction

Maceration and infusion are relatively simple methods for preliminary flower extraction. In maceration, powdered or cut plant material is contacted with a selected solvent for a defined period, followed by filtration and concentration. Infusion uses hot or warm water and may better represent beverage-type preparations. Prolonged heating should be avoided unless stability has been evaluated. Soxhlet extraction may be useful for selected less-polar constituents but is not automatically suitable for heat-sensitive anthocyanins.

5.2 Ultrasound-assisted extraction (UAE)

UAE uses acoustic cavitation to improve solvent penetration and mass transfer. It may reduce extraction time and solvent consumption. However, amplitude or power, frequency, temperature, time and solvent

composition must be controlled because excessive energy input and heating may affect pigment stability.

5.3 Microwave-assisted extraction (MAE)

MAE uses microwave energy to heat polar solvents and moisture within plant tissues, potentially improving extraction efficiency. Optimization should include microwave power, time, solvent composition, sample-to-solvent ratio and temperature. A higher extraction yield should not be assumed to mean a better-quality extract unless the marker profile and degradation products are also assessed.

5.4 Supercritical-fluid extraction

Supercritical carbon dioxide is more suitable for non-polar or lipophilic compounds unless a suitable polar modifier is used. It should not be presented as the default method for extracting highly polar ternatins. Its relevance to C. ternatea depends on the target fraction, process design and supporting experimental evidence.

5.5 General extraction workflow

Step	Process / control point
1	Authenticate and document the flower material
2	Clean, dry under defined conditions and reduce particle size
3	Select water or aqueous ethanol according to the target profile
4	Control solvent ratio, pH, time and temperature
5	Filter and concentrate under conditions that limit degradation
6	Record extract yield and retain a representative sample
7	Perform fingerprinting, marker analysis and stability testing

6. ANALYTICAL METHODS FOR QUALITY ASSESSMENT

A combination of identity tests and chemical assays is preferable to reliance on a single test. Analytical methods should be selected according to the purpose: preliminary screening, fingerprint comparison, marker quantification or structural characterization.

6.1 Thin-layer chromatography (TLC) and HPTLC

TLC can provide a preliminary fingerprint and help compare batches under the same conditions. HPTLC offers improved resolution and documentation and may support semi-quantitative or quantitative analysis when a suitable reference standard and validated procedure are available. The mobile phase, stationary phase, application volume, chamber saturation and detection conditions should be reported.

6.2 High-performance liquid chromatography (HPLC)

HPLC can separate and quantify selected anthocyanin or flavonoid markers. For ternatin-rich extracts, the method should be optimized for the chemical characteristics of the target compounds. Method validation should address specificity, linearity, range, accuracy, precision, robustness and solution stability. Reference standards and clear peak-identification criteria are important.

6.3 LC–MS and structural characterization

Liquid chromatography coupled with mass spectrometry can provide information about molecular mass and fragmentation patterns and is useful for profiling complex mixtures. NMR may be used when isolation and detailed structural elucidation are required. These techniques provide stronger characterization than a nonspecific total-content assay but require appropriate instrumentation and expertise.

6.4 UV–visible spectroscopy

UV–visible spectroscopy can monitor the visible absorption behaviour of flower pigments and compare colour changes under controlled pH conditions. It is useful for screening and stability studies, but absorbance at a selected wavelength alone cannot confirm the identity or purity of individual ternatins.

Method	Main purpose	Important limitation
TLC	Preliminary fingerprint	Limited quantification and resolution
HPTLC	Batch comparison and fingerprint documentation	Requires method optimization and suitable standards
HPLC	Separation and marker quantification	Needs validated method and reference material
LC–MS	Molecular profiling and characterization	Cost, expertise and complex data interpretation
UV–visible	Pigment absorbance and colour behaviour	Not specific for individual compounds
NMR	Structural elucidation of isolated compounds	Requires sufficient purified material

7. STANDARDIZATION AND QUALITY CONTROL

Standardization should connect the identity of the raw plant material with the chemical and physical quality of the finished extract. A suitable specification may include botanical identity, plant part, source, foreign matter, moisture, ash values, extractive values, extraction yield, chromatographic fingerprint, marker content and relevant contaminant limits.

7.1 Suggested quality-control framework

For a flower extract, a practical development framework is to define the botanical source and flower colour; establish drying and extraction parameters; select a chromatographic fingerprint; quantify one or more suitable marker compounds when reference standards are available; and assess batch-to-batch consistency. Total anthocyanin content may be included as a supportive test, but it should not replace marker-specific profiling.

7.2 Contaminant and microbiological testing

Depending on the intended product and applicable standards, testing may include microbial limits, pesticide residues, heavy metals, mycotoxins, foreign matter and residual solvents. Good agricultural and collection practices and good manufacturing practices can help reduce variability and contamination. The applicable limits must be taken from the relevant current regulatory or compendial source.

7.3 Stability-indicating quality control

A stability-indicating approach should monitor changes in marker content and chromatographic profile during storage. Packaging, temperature, light, oxygen, humidity and pH may influence pigment stability. The storage condition and sampling schedule should be justified for the intended product.

8. FORMULATION AND PIGMENT STABILITY

Ternatin-rich extracts are of interest as natural colour ingredients in beverages and other food systems. Their colour may shift with pH, while heat, light, oxygen, metal ions and formulation ingredients can affect stability. A formulation study should therefore evaluate the actual product matrix rather than relying only on results from a simple aqueous solution.

8.1 Formulation considerations

For a pharmaceutical or nutraceutical preparation, the intended route, dose, excipients, pH, packaging and storage conditions should be defined before selecting an extract. Compatibility studies should examine whether excipients alter colour, marker recovery, precipitation or degradation. A food colourant application and a medicinal formulation have different specifications and regulatory requirements.

8.2 Encapsulation and protection strategies

Microencapsulation and other protective delivery approaches have been explored for anthocyanin-rich materials. Such strategies may improve handling or protect pigments under selected conditions, but improved physical stability does not automatically demonstrate improved absorption or therapeutic performance. The encapsulated product should be assessed for marker recovery, release behaviour, moisture, particle properties and storage stability.

8.3 Packaging and storage

Light-resistant packaging and controlled storage may be appropriate where photodegradation or moisture uptake is a concern. The final storage recommendation should be based on stability data for the specific extract or formulation, not copied from another plant product.

9. SAFETY AND REGULATORY CONSIDERATIONS

Safety assessment is product-specific. A traditional flower infusion, concentrated hydroalcoholic extract and purified peptide are not interchangeable. Traditional use alone cannot establish the safety of concentrated extracts, high doses or long-term use. Relevant safety studies should consider the plant part, extraction solvent, dose, route, duration, contaminants and intended population.

Regulatory requirements depend on whether the product is developed as a food ingredient, colour additive, cosmetic, traditional medicine, nutraceutical or pharmaceutical product. Each category has different requirements for identity, purity, claims, safety and manufacturing quality. Current local rules should be checked before making a submission or commercial claim.

10. CHALLENGES AND RESEARCH GAPS

A major challenge is the lack of uniformity between published preparations. Differences in cultivar, flower colour, plant maturity, drying, extraction solvent and analytical methods make direct comparison difficult. Some studies report total phenolic or antioxidant values without adequate chemical characterization. Such results are useful for preliminary screening but do not provide a complete quality specification.

Further work should prioritize authenticated raw material, reference standards for selected ternatins, validated HPLC or HPTLC methods, stability-indicating assays and inter-batch reproducibility. Comparative studies of UAE, MAE and conventional extraction should report both extraction yield and marker recovery. Formulation studies should use real product matrices and include appropriate storage conditions.

11. FUTURE PERSPECTIVES

Future development should move from general phytochemical screening toward reproducible, marker-defined extracts. A shared minimum reporting framework could include botanical voucher information, plant part, flower colour, harvest and drying details, solvent composition, extraction ratio, yield, chromatographic fingerprint, marker concentration and stability data.

The use of green extraction technologies may be valuable if they demonstrate reproducible marker recovery, reduced solvent use and acceptable scale-up performance. Digital image-based colour measurement and chemometric analysis may complement chromatographic quality control, provided that the methods are validated. More work is also needed on reference materials, long-term stability and product-specific safety.

CONCLUSION

Clitoria ternatea L. is a promising botanical source of distinctive flower pigments and other phytochemical constituents. Ternatins are important markers for flower-extract characterization, while flavonols, phenolic acids and tissue-specific constituents contribute to the wider chemical profile. The quality of a C. ternatea preparation depends on correct botanical authentication, controlled collection and processing, suitable extraction, reliable analytical characterization and appropriate storage.

TLC and HPTLC can support fingerprinting, HPLC and LC–MS can provide more detailed marker analysis, and UV–visible spectroscopy can monitor pigment behaviour. No single method is sufficient for all quality questions. Standardized reference materials, validated analytical procedures, batch-to-batch assessment and formulation-specific stability and safety data are necessary to support reproducible pharmaceutical development. The present review provides a quality-focused framework and does not claim new experimental results or established clinical efficacy.

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