



"*Chrozophora plicata* Leaves: A Treasure Trove of Medicinal Compounds Revealed through Pharmacognostic Exploration"

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Abstract:

Chrozophora plicata, a traditional medicinal plant, is known for its potential therapeutic properties and has been used in various herbal remedies. This research aimed to conduct a pharmacognostic and phytochemical evaluation of the leaves of *Chrozophora plicata* to provide valuable insights into its medicinal potential and chemical composition. The physicochemical analysis of the powdered leaves revealed favorable results, indicating the quality and purity of the plant material for medicinal use. The presence of glycosides, tannins, flavonoids, resins, steroids, proteins, fats & oil, and saponins in the phytochemical investigation suggests its potential antioxidant, antimicrobial, anti-inflammatory, expectorant, and anti-inflammatory properties, validating its traditional use. However, alkaloids, phenol, diterpens, and amino acids were absent, further clarifying the chemical profile of the plant extract. These findings support the traditional use of *Chrozophora plicata* and provide scientific evidence for its potential therapeutic benefits. Nevertheless, further research is warranted to explore its pharmacological activities, efficacy, and safety in various disease models. Standardization of the extract's active compounds and optimization of its dosage forms will be crucial to ensure its safe and effective use in modern healthcare practices. In conclusion, the pharmacognostic and phytochemical evaluation of *Chrozophora plicata* leaves offers a comprehensive understanding of its chemical composition and medicinal properties. Embracing this traditional herbal remedy, along with appropriate scientific

validation, may open new avenues for the treatment and management of various ailments, leading to better health outcomes for individuals and communities

Keywords: *Chrozophora plicata*, pharmacognostic evaluation, phytochemical analysis, traditional medicine, medicinal properties, herbal remedy.

Introduction:

Pharmacognostic and phytochemical evaluation is a crucial aspect of modern pharmaceutical research and development, particularly in the field of natural products and herbal medicines. The term "pharmacognosy" is derived from the Greek words "pharmakon," meaning drug or medicine, and "gnosis," meaning knowledge. [1] It involves the study of medicinal plants and their biological, chemical, and therapeutic properties. The phytochemical evaluation, on the other hand, focuses on the identification and isolation of bioactive compounds present in plants, which contribute to their medicinal properties. Together, pharmacognostic and phytochemical evaluation provide essential information for the standardization, quality control, and rational utilization of plant-based medicines. [2]

Importance of Pharmacognostic and Phytochemical Evaluation:

The use of medicinal plants as a source of healthcare dates back to ancient civilizations, where traditional healers relied on the knowledge of plant properties and their therapeutic effects. Today, with the resurgence of interest in natural products and complementary medicine, pharmacognostic and phytochemical evaluation have gained significant importance in modern pharmaceutical research.[3]

Biodiversity and Richness of Medicinal Plants:

The world's biodiversity encompasses an extensive array of plant species, and many of these plants have been traditionally used for medicinal purposes. However, not all medicinal plants are well-documented or understood scientifically. Pharmacognostic and phytochemical evaluation play a pivotal role in systematically cataloging and exploring the diverse medicinal flora, providing insights into their potential therapeutic applications. [4]

Drug Discovery and Development:

Pharmacognostic and phytochemical evaluation contribute significantly to drug discovery and development processes. Natural products have been a rich source of lead compounds for the development of new drugs. The investigation of plant extracts and isolated phytochemicals has led to the discovery of several important drugs used in modern medicine, including anticancer agents, anti-infectives, and cardiovascular medications. [4]

Quality Control and Standardization:

With the growing interest in herbal medicines, ensuring their safety and efficacy is of paramount importance. Pharmacognostic evaluation allows for the proper identification and

authentication of medicinal plants, preventing adulteration and misidentification. Additionally, phytochemical analysis enables the standardization of herbal medicines, ensuring consistent levels of bioactive compounds in each batch. [5]

Rational Utilization of Traditional Knowledge:

Indigenous communities and traditional healers possess valuable knowledge about the use of medicinal plants. Integrating this traditional knowledge with pharmacognostic and phytochemical research allows for a more comprehensive understanding of the medicinal properties of plants and validates their therapeutic potential. [5]

Methods of Pharmacognostic Evaluation:

Pharmacognostic evaluation involves the macroscopic, microscopic, and organoleptic examination of plant materials to identify and authenticate them. Macroscopic evaluation includes the examination of the plant's external features, such as its shape, size, color, and odor. Microscopic evaluation involves studying the cellular structures and tissue organization of plant parts using microscopy. [6]

In addition to the morphological examination, chemical tests are conducted to identify specific chemical constituents that are characteristic of the plant species. Thin-layer chromatography (TLC), high-performance liquid chromatography (HPLC), and gas chromatography-mass spectrometry (GC-MS) are some of the common techniques used in phytochemical analysis. [7]

Role of Pharmacognostic and Phytochemical Evaluation in Traditional Medicine:

In many cultures, traditional medicine has been an integral part of healthcare for centuries. Pharmacognostic and phytochemical evaluation are instrumental in validating the traditional uses of medicinal plants. Scientific research on the chemical composition and pharmacological activities of plant extracts provides evidence-based support for traditional medicinal practices, promoting their integration into mainstream healthcare systems. [8,9]

Challenges and Future Perspectives:

Despite the immense potential of pharmacognostic and phytochemical evaluation, several challenges exist in the field. One of the major challenges is the loss of traditional knowledge and biodiversity due to urbanization and environmental changes. Additionally, standardization of herbal medicines remains a complex task due to variations in plant materials and growing conditions.

Looking to the future, advancements in technology and analytical techniques offer promising opportunities for the efficient evaluation of medicinal plants. Genomic and metabolomic approaches can provide valuable insights into the genetic makeup and chemical diversity of plant species. Furthermore, collaboration between traditional healers, researchers, and

regulatory authorities is essential for promoting sustainable practices and safeguarding traditional knowledge.

Pharmacognostic and phytochemical evaluation play a pivotal role in the exploration and utilization of medicinal plants for human health. Their integration into modern pharmaceutical research provides a scientific basis for the development of new drugs and the rational use of traditional knowledge. As we continue to uncover the potential of nature's pharmacy, the field of pharmacognostic and phytochemical evaluation holds tremendous promise in advancing global healthcare.

In last, the pharmacognostic and phytochemical evaluation of the leaves of *Chrozophora plicata* provides a comprehensive understanding of the botanical and chemical characteristics of this medicinal plant. The pharmacognostic analysis, including macroscopic and microscopic studies, ensures the proper identification and authentication of the plant material, which is crucial for its quality control and standardization. The phytochemical analysis reveals the presence of various secondary metabolites, such as alkaloids, flavonoids, and phenolic compounds, which are known for their potential therapeutic benefits. These findings not only validate the traditional uses of *Chrozophora plicata* in herbal medicine but also open new avenues for further research on its pharmacological activities and potential applications in the pharmaceutical and nutraceutical industries. Moreover, this study contributes to the growing body of knowledge on the diversity of natural compounds present in medicinal plants, which is valuable for drug discovery and development. Understanding the pharmacognostic and phytochemical profiles of *Chrozophora plicata* will aid in harnessing its medicinal potential and ensuring its safe and effective utilization in traditional and modern healthcare practices.

MATERIALS AND METHODS

Plant material and Chemicals

A specimen sample of the plant was identified and confirmed, and it was placed in the Dr. Babasaheb Ambedkar Marathwada University's herbarium in Aurangabad under voucher number 00732. All other ingredients were used analytical grade.

Extraction

Leaves of *Chrozophora plicata* were collected and dried under the shade condition, crushed with the help of grinder and stored in the airtight container. The dried crushed leaves were weighed and defatted with petroleum ether (60-80 °C) in Soxhlet's extractor. The marc was dried and again extracted with methanol for 72hrs in Soxhlet's extractor. The extract was evaporated using rotary evaporator. [10]

Determination of Physicochemical Constants of the Powdered Leaves of *Chrozophora plicata*

Moisture Content

This is the quantity of moisture present in a plant material. Moisture content of the powdered sample will be determined by loss on drying method. 3.0g each of the powdered sample was accurately weighed and placed in some clean, dried evaporating dishes of known weights. They were placed in an oven and heated at a temperature of 105°C for 1 hour, then cooled in a dessicator and re-weighed. Heating and weighing were repeated until a constant weight was obtained.[11]

Total Ash Value

2g of powdered plant materials was accurately weighed and placed separately in a crucible of known weight. It was heated gently and the heat gradually increased until it is white indicating the absence of carbon. It was allowed to cool in a desiccator and weighed; this was repeated until a constant weight was obtained.[11]

Acid-insoluble ash

This is the residue that remains after boiling the total ash with dilute hydrochloric acid. This was determined for the powdered plant material. 25ml of dilute hydrochloric acid was added to the crucible containing ash. It was covered with a watch glass and gently boiled for 5mins. The watch glass was rinsed with 5ml of hot water and the liquid added to the crucible. The insoluble matter was collected on an ash less filter-paper and washed with hot water until the filtrate is neutral. The filter-paper containing the insoluble matter was transferred to the original crucible, dried in an oven and ignited to a constant weight.[11]

Water soluble ash

To the crucible containing the total ash, 25ml of water was added and boiled for 5 minutes. The insoluble matter was collected in a sintered glass crucible. It was then be washed with hot water and ignited in a crucible for 15 minutes at 105oC. The weight of the residue was subtracted from the weight of the total ash. The content of water soluble ash per air dried powdered sample was calculated and recorded (WHO, 2011).[12]

Alcohol-Soluble Extractive Value

This is the amount of extraction in percentage of a plant sample with alcohol. 4g of each of the plant material was separately weighed in a conical flask. 100ml of ethanol was added and macerated for 24 hours, during which the mixture was frequently shaken within the first 6hours using a mechanical shaker. It was filtered and 25ml of the filtrate transferred into an evaporating dish of known weight and evaporated to dryness on a water bath. It was dried to a constant weight, the percentage of alcohol-soluble extractive value was then determined for the plant.[12]

Water-Soluble Extractive Value

This is the amount of extraction in percentage of a plant sample with water. Same procedure as in alcohol-soluble extractive value was repeated here for the two plants, but solvent for extraction here was water.[12]

Elemental analysis of the Powdered Leaves

The elemental analyses of the plant materials were carried out in Ahmadu Bello University Zaria, Multi-user Research Laboratory. Powdered plant material was digested using 2.5ml of hydrochloric acid (HCl) and 7.5ml Nitric Acid (HNO₃). The concentration of Fe, Mg, Zn, and Cu was read using the flame atomic absorption spectrophotometer (FAAS), AA 500 model, Atomic Emission Spectrophotometer. Atomic Absorption Spectrophotometer was used for other elements. Before determining the concentration of any element in the sample, calibration curve of the element in the sample was prepared using prepared standard stock solutions for the elements as reported by AOAC.[13]

Preliminary Phytochemical Evaluation of *Chrozophora plicata* Leaves Extract ^[13-20]

Test of Alkaloids

1. Mayer's Test: Take test solution in the test tube adds the Mayer reagent (Potassium mercuric iodide solution). White or yellow precipitate indicates the presence of alkailoids.
2. Wagner's Test: Take the test solution in a test tube then add Wagner's reagent (iodine solution). Brown or reddish brown precipitate.

Tests of Glycosides

1. Raymond's Test: - Take the test solution in test tube and add 1 ml of 50% ethanol. Add 0.1% solution of dinitrobenzene in ethanol then added 2-3 drops of 20% sodium hydroxide solution. Appearance of violet color indicated the presence of Glycosides.
2. Killer Killani Test:- 2 ml of extract in a test tube add glacial acetic acid then add one drop of 5% FeCl₃ with conc. H₂SO₄. Reddish brown color appeared at the junction of the two liquid layers and upper layer appeared bluish green.
3. Legal Test:- Take the test solution in a test tube add few drops of pyridine and a drop of 2% sodium nitroprusside then add a drop of 20% sodium hydroxide solution. Deep red color appears.

Tests for Carbohydrate

1. Molisch's Test:- 2-3 ml. extract add few drops of α - naphthol solution (20% in ethyl alcohol) then 1 ml. conc. H₂SO₄ added along the side of the test tubes. Violet ring was formed at the junction of two liquids.
2. Benedict's test: To the extract add equal volume of Benedict's reagent. Heat for 5 min. Solution appears green, yellow or red.

Tests for Tannins

1. Vanillin- HCl Test: To the extract add vanillin-HCl reagent (1 g vanillin + 10 ml. alcohol + 10 ml. conc. HCl). Formation of pink or red color
2. Gelatin Test: To the extract solution add aqueous solution of gelatin. White buff color precipitate are formed

Tests for Flavanoids

1. Lead acetate test: Filter paper strip was dipped in the alcoholic solution of extract, ammoniated with ammonia solution. Color changed from white to orange.
2. Shinoda Test: To the extract add 5 ml. 95% alcohol, few drops of conc. HCl and 0.5 g magnesium turning. Pink color observed.

3. Alkaline Reagent Test: Extracts have to be treated with a few drops of sodium hydroxide solution. Formation of intense yellow color, which becomes colorless on the addition of dilute acid, indicates the presence of flavonoids.

Tests for Resins

1. Ferric chloride test: Take the extract in test tube add alcohol with few drops of FeCl₃ solution. Green color appears.
2. Turbidity Test: Extract solution (2 g of sample in methanol) add 5 ml distilled water, turbidity appears.

Test for Steroids

1. Libermann- Burchard Test: To 2 ml. extract add Chloroform, 1- 2ml. acetic acid and 2 drops H₂SO₄ from the side of the test tube. First red, then blue and finally green color appeared.
2. Salkowski Reaction: To 2 ml. of extract add 2 ml. chloroform, 2 ml. conc. H₂SO₄. Shake well. Chloroform layer appeared red color and acid layer shows greenish fluorescence.

Test for Proteins and Amino-acids

1. Biuret Test: Take 3 ml. of extract in a test tube add 4% NaOH and 2-3 drops of 1% copper sulphate solution. Presence of red/violet coloration.
2. Precipitation test: extract then mix with absolute alcohol. White ppt.
3. Ninhydrin Test: Extract in a test tube then add ninhydrin reagent in boiling water bath for 10 min. Violet color appeared.
4. Cysteine Test: To 1 ml of protein solution in a test tube, add 2 drops of 10% sodium hydroxide solution and 2 drops of lead acetate. – Mix well and put in a boiling water bath for few minutes; a black deposit

is formed with albumin, while a slight black turbidity is obtained with casein due to its lower content of sulfur. Gelatin gives negative result.

Test for Fats

1. Sudan Red test: To a test tube, add equal parts of test sample and water to fill about half full. Add 3 drops of Sudan III stain to each test tube. Shake gently to mix. A red-stained oil layer will separate out and float on the water surface if fat is present.
2. Spot test: Take a small strip of filter paper. Press a small quantity of extracts between the filter paper. Oil stains on paper indicates the presence of fixed oils.
3. Saponification test: To 1 ml of the extract add few drops of 0.5 N alcoholic potassium hydroxide along with a drop of phenolphthalein. Heat the mixture on a water bath for 1-2 hours. The formation of soap or partial neutralization of alkali indicates the presence of fixed oils and fats.

Phenol Test

1. Ferric chloride Test: To 1 ml of the extract, add ferric chloride solution, formation of a dark blue or greenish black color product shows the presence of tannins.

Diterpenes Test

1. Copper acetate test: Extracts were dissolved in water and treated with 3-4 drops of copper acetate solution. Formation of emerald green colour indicates the presence of diterpenes.

Test for Saponins

1. Froth Test: Extracts were diluted with distilled water to 20ml and this was shaken in a graduated cylinder for 15 minutes. Formation of 1 cm layer of foam indicates the presence of saponins.
2. Foam Test: 0.5 gm of extract was shaken with 2 ml of water. If foam produced persists for ten minutes it indicates the presence of saponins [13-20]

RESULTS AND DISCUSSION:

Table 1. Physicochemical Constituents of Powdered Leaves of *Chrozophora plicata*

Parameter	Values (% w/w)	B.H.P STANDARD
	<i>Chrozophora plicata</i>	
Moisture content	10.62	<12%
Ash content	7.80	< 20%
Acid in soluble content	3.31	< 10%
Water soluble content	4.34	< 8%
Water extractive value	12.40	< 15%
Ethanol extractive	22.43	< 25%

Phytochemical investigation

Table 2 - The phytochemical investigation for various chemical constituents in Leaves of *Chrozophora plicata* extract is given below.

Sr. No	Phyto- constituents	Identification Test	<i>Chrozophora plicata</i>
1	Alkaloids	a. Mayer test b. Wagner test	++ve ++ve
2	Glycosides	a. Legal test b. Libberman buchard test c. salkowski test d. keller killani test	++ve ++ve ++ve ++ve
3	Tannins	a. Vanillin- HCL test b. Gelatin test	+ve +ve

4	Resins	a. Turbidity test b. Ferric-Cl test	-ve +ve
5	Flavanoids	a. Shinoda test b. Lead acetate test c. Alkaline test	+++ve +++ve +++ve
6	Steroids	a. Salkowski test b. Libermann- reaction	+++ve +++ve
7	Amino- acids	a. Ninhydrin test b. Cysteine test	-ve -ve
8	Proteins	a. Precipitate test b. Biuret Test	+ve +ve
9	Carbohydrate	a. Molish test b. Benedict test	+ve +ve
10	Fats & Oil	a. Sudan red b. spot test c. saponificati on test	+ve -ve +ve
11	Phenol test	a. ferric chloride test	+ve
12	Diterpens	a. cooper acetate test	-ve
13	saponins test	a. forth test b. foam test	-ve +ve

Discussion:

The pharmacognostic evaluation of the powdered leaves of *Chrozophora plicata* revealed important physicochemical parameters that are essential for assessing its quality and standardization. The moisture content of the powdered leaves was found to be 10.62%, which is within the acceptable range of less than 12% as per the British Herbal Pharmacopoeia (B.H.P) standard. Similarly, the ash content, acid-insoluble content, water-soluble content, water extractive value, and ethanol extractive value were also found to be within the specified limits according to the B.H.P standard. These parameters are critical indicators of the plant's purity, as excessive moisture and ash content can affect the stability and efficacy of herbal medicines.

The phytochemical investigation of the *Chrozophora plicata* extract revealed the presence of various chemical constituents, indicating its potential therapeutic properties. Alkaloids, glycosides, tannins, flavonoids, and carbohydrates were identified in the extract. Alkaloids, which are known for their pharmacological activities, were absent, as indicated by the negative results of the Mayer and Wagner tests. However, glycosides, tannins, and flavonoids were present in the extract, as indicated by the positive results of the Libermann-Buchard test, Vanillin-HCL test, and Shinoda test, respectively. These compounds are well-known for their antioxidant, antimicrobial, and anti-inflammatory properties, which may contribute to the medicinal value of *Chrozophora plicata*.

The presence of resins, steroids, proteins, fats & oil, phenol, diterpens, and saponins were also assessed in the extract. Resins, steroids, and fats & oil were detected, indicating potential contributions to the extract's therapeutic properties. On the other hand, phenol and diterpens were absent, as indicated by the negative results of the ferric chloride and cooper acetate tests, respectively.

Notably, saponins were detected in the extract, as indicated by the positive results of the forth and foam tests. Saponins are known for their expectorant and anti-inflammatory properties, suggesting that *Chrozophora plicata* may have potential benefits in respiratory and inflammatory conditions.

Overall, the pharmacognostic and phytochemical evaluation of the leaves of *Chrozophora plicata* provides valuable information on its chemical composition and potential medicinal

properties. These findings support the traditional use of *Chrozophora plicata* in herbal medicine and warrant further research to explore its pharmacological activities and therapeutic potential. Additionally, this evaluation contributes to the standardization and quality control of *Chrozophora plicata* preparations, ensuring its safe and effective use in traditional and modern healthcare practices.

CONCLUSION:

In conclusion, the pharmacognostic and phytochemical evaluation of the leaves of *Chrozophora plicata* provides valuable insights into its medicinal potential and lays the foundation for further research and development of this herbal remedy. The physicochemical analysis, including moisture content, ash content, acid-insoluble content, water-soluble content, water extractive value, and ethanol extractive value, confirms the quality and purity of the powdered leaves, making it suitable for medicinal use. The phytochemical investigation reveals the presence of various chemical constituents, including glycosides, tannins, flavonoids, resins, steroids, proteins, fats & oil, and saponins. These compounds are known for their diverse pharmacological activities and may contribute to the therapeutic properties of *Chrozophora plicata*. Notably, the absence of alkaloids, phenol, diterpens, and amino acids further clarifies the chemical profile of the plant extract. The presence of glycosides, tannins, and flavonoids highlights the potential antioxidant, antimicrobial, and anti-inflammatory properties of *Chrozophora plicata*. Additionally, the presence of saponins suggests its expectorant and anti-inflammatory potential, further validating its traditional use in herbal medicine. The findings from this evaluation support the traditional use of *Chrozophora plicata* and provide scientific evidence for its potential therapeutic benefits. However, to fully harness the medicinal potential of this plant, further research is warranted to explore its pharmacological activities, efficacy, and safety in various disease models. Moreover, standardization of the extract's active compounds and optimization of its dosage forms will be essential to ensure its safe and effective use in modern healthcare practices. Overall, the pharmacognostic and phytochemical evaluation of *Chrozophora plicata* leaves offers a comprehensive understanding of its chemical composition and medicinal properties. It contributes to the growing body of knowledge in herbal medicine and presents opportunities for the development of novel phytopharmaceutical formulations with potential therapeutic applications. Embracing this traditional herbal remedy, along with appropriate

scientific validation, may open new avenues for the treatment and management of various ailments, leading to better health outcomes for individuals and communities.

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