

Ethnobotanical, Phytochemical And Pharmacological Screening Of Some Regional Plants

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ABSTRACT:

Introduction As we know that India is one of the country in the world who using ayurvedic drugs in routing life, so the purpose of this study is only to introduced traditional drugs for prevention of life of the patient. As I selected four plant *Artemisia Japonica*, *Martynia annua*, *Erythrina variegata*, *Jatropha curca* for the work. **Objectives of the Proposed Study** Isolation of Bioactive from *Artemisia Japonica*, *Martynia annua*. Pharmacognostic studies of *Artemisia Japonica*, *Martynia annua*. To study Antibacterial, Anti-inflammatory, antioxidant & anticancer activity. **Material and Method** Isolation of Bioactives done by using Continuous Hot Extraction with ethyl alcohol as solvent. **Result & Discussion** Pharmacognostic evaluation of the plant was carried out through macroscopic and microscopic studies, while antimicrobial activity was assessed by the zone of inhibition method against two bacterial strains; cytotoxicity was evaluated using the brine shrimp lethality assay, and antioxidant activity was also determined. Conclusion as per the result and discussion we conclude that the plant first *Artemisia Japonica* shows promising result as compare to other plants.

Key words: Alkaloids, Asteraceae, Euphorbiaceae, Martyniaceae, Moraceae.

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INTRODUCTION:

Ethnobotany encompasses the study of the intricate relationship between humans and plants. The reliance of human societies on plants dates back approximately 10,000 years, beginning with domestication. Ethnobotanical research documents traditional knowledge regarding the cultural interactions between people and plants, exploring how local communities have historically utilized plants for diverse purposes, including their integration into cultural traditions and religious practices [1]. In many developing nations, ethnomedicinal knowledge remains a primary source of healthcare, as modern medical systems often face challenges such as inadequate staffing and the high cost of pharmaceutical drugs. Ethnomedicine is rooted in folk beliefs, traditional knowledge, and practices used to maintain health and treat diseases. However, this valuable knowledge is rapidly disappearing due to socio-economic, environmental, and technological transformations. Therefore, systematic documentation and conservation efforts are crucial to preserving ethnomedicinal wisdom before it is lost forever [2].

Scope of Ethnobotany

Ethnobotany is a multidisciplinary field that integrates knowledge from various disciplines, including botany, anthropology, ecology, linguistics, economics, and ethnopharmacology. As a subset of ethnobiology, it primarily focuses on the relationship between humans

and plants, given their significance in food systems, traditional medicine, and cultural practices. Historically, ethnobotany combined botanical and ethnological interests, approached from both practical and philosophical perspectives. Professionals in the field contribute from different angles—botanists study economic plant benefits, anthropologists explore traditional plant management, and ecologists examine interactions between societies and plant resources. Ethnobotanical research plays a vital role in modernizing traditional agricultural systems, industrialization, food security, conservation, and the documentation of indigenous knowledge [3,4].

The field is categorized into four major areas: (1) basic ethnobotany, which documents traditional plant knowledge; (2) quantitative ethnobotany, which evaluates plant-use patterns; (3) experimental ethnobotany, which tests hypotheses and predicts outcomes; and (4) applied ethnobotany, which focuses on practical applications like pharmaceutical research and conservation. These interconnected fields contribute to preserving and utilizing traditional botanical knowledge in contemporary contexts [5].

Importance of Ethnobotany

Ethnobotany plays a crucial role in understanding the relationship between humans and plants, encompassing their cultural significance, ecological roles, and practical applications. It contributes to the

documentation of traditional botanical knowledge, indigenous food production, and the scientific evaluation of medicinal plants. Additionally, ethnobotany explains biodiversity, cultural diversity, and traditional resource management practices. The field highlights the cultural importance of plants in providing food, shelter, clothing, tools, and ceremonial uses. It also explores the distribution of plants and the transmission of botanical knowledge across generations. Modern ethnobotanical research continues to inspire innovations in plant-based production and the enhancement of traditional manufacturing methods [6,7].

Importance of Traditional Knowledge in Medicinal Plants

Indigenous knowledge refers to the accumulated wisdom, skills, and practices developed by local communities over generations through observation and experience. This knowledge plays a crucial role in the use, management, and conservation of medicinal plants, which are essential for food, healthcare, and daily life. Across the world, traditional communities possess extensive botanical knowledge, ensuring the sustainable use of plant resources. Sustainable development researchers recognize indigenous knowledge for its valuable insights into resource management, ecological practices, and the relationship between communities and nature. According to the Convention on Biological Diversity (CBD), this knowledge is shaped by cultural heritage and long-term territorial presence. It is often preserved through oral traditions, passed down from one generation to the next. Traditional knowledge is deeply embedded in the cultural identity of a community, influencing belief systems, healthcare, and environmental conservation. Over time, these practices evolve, adapting to changing environments while maintaining their core significance. The widespread use of traditional medicine in many countries highlights the enduring value of indigenous knowledge in promoting health and sustainability [8].

Importance of Medicinal Plants

a) Plants Used in Human Health

In resource-poor communities, traditional medicinal plants remain the most accessible and affordable form of healthcare. In Ethiopia, as in many developing countries, modern healthcare services are often inadequate, unavailable, or out of reach for most people. Due to a rapidly growing population and cultural resistance to modern medicine, approximately 80% of Ethiopians rely on traditional medicine for healthcare, with over 90% of livestock treatments also

depending on medicinal plants. Traditional medicine is preferred due to its accessibility, affordability, and perceived effectiveness. The widespread use of medicinal plants in Ethiopia is driven by socio-cultural acceptance, availability, and their biomedical benefits, making traditional medicine the most widely accepted alternative healthcare system across the country [9].

Plants used in Ethnoveterinary

In Ethiopia and other developing countries, livestock diseases significantly impact animal productivity. Due to limited access to modern veterinary medicine, traditional medicinal plants play a crucial role in managing livestock health. Ethiopian communities possess rich ethnoveterinary knowledge and others to treat livestock ailments. Ethnoveterinary medicine includes herbal remedies, traditional surgeries, immunization, and cultural healing practices. Despite Ethiopia having one of Africa's largest livestock populations, ethnoveterinary knowledge remains largely undocumented. Raising awareness and systematically documenting these practices are essential for preserving valuable livestock healthcare traditions and improving animal health management [10].

Importance of Documenting Ethnopharmacological Information

Indigenous communities possess valuable knowledge of natural resources, particularly medicinal plants. However, this knowledge is rapidly declining due to deforestation, modernization, and cultural shifts. Less than 1% of indigenous cultures have been studied for their medicinal plant knowledge, and in industrialized regions, it is disappearing due to urbanization and cultural assimilation. In Ethiopia, traditional medicinal knowledge is unevenly distributed and passed down orally, making it vulnerable to loss due to displacement, modernization, and generational neglect. Preserving this knowledge is crucial, requiring systematic documentation of medicinal plants and their uses. Such efforts not only protect cultural heritage but also support sustainable development and provide economic opportunities for local communities [11].

Ethnopharmacological Approach in Plant-based Drug Discovery

Drug discovery, especially for ailments lacking effective medication, relies on extensive research and development. Screening methods to identify plant-based compounds can be time-consuming, costly, and inefficient. High-throughput screening, genomics, and combinatorial chemistry offer potential improvements. Ethnopharmacology, an interdisciplinary field, plays a vital role by investigating traditional folk remedies to

discover active plant components with therapeutic potential. This approach considers plants' biological activity and involves preclinical studies, isolation of active compounds, and structure-activity relationship analysis. Despite challenges such as variability in raw materials and regulatory complexities, natural products continue to contribute to drug discovery, especially in developing countries with rich traditional knowledge. Modern methods like reverse ethnopharmacology link traditional uses with biomedical applications. However, issues such as standardization, ethical concerns, and potential plant-drug interactions require further research to ensure safe and effective use. Standardization, based on chemical profiling or marker compounds, is essential for reproducibility. The use of herbal medicines is increasing, but interactions with conventional drugs are a growing concern. Continued research is necessary to fully understand these interactions and ensure the safe integration of plant-based remedies with modern pharmaceuticals [12].

PHYTOCHEMISTRY: LINKING TRADITION AND SCIENCE

Phytochemicals, derived from the Greek word *phyto* (meaning plant), are naturally occurring bioactive compounds found in plants. They offer health benefits beyond those provided by macronutrients and micronutrients. These compounds play a crucial role in protecting plants from diseases, environmental stressors, and damage while also contributing to their color, aroma, and flavor. Phytochemicals help safeguard plant cells against various environmental hazards, including pollution, stress, drought, UV radiation, and pathogenic attacks. Recent research has established their significant role in promoting human health, particularly when consumed in adequate amounts. Over 4,000 phytochemicals have been identified and classified based on their protective functions, physical properties, and chemical characteristics, with approximately 150 studied in detail.

Phytochemicals are widely distributed across various dietary sources, including fruits, vegetables, legumes, whole grains, nuts, seeds, fungi, herbs, and spices. Common sources include broccoli, cabbage, carrots, onions, garlic, whole wheat bread, tomatoes, grapes, cherries, strawberries, raspberries, beans, legumes, and soy-based foods. These bioactive compounds accumulate in different parts of plants, such as the roots, stems, leaves, flowers, fruits, and seeds. Many phytochemicals, especially pigment molecules, are highly concentrated in the outer layers of plant tissues. Their levels vary depending on factors such as plant

variety, cultivation conditions, processing, and cooking methods. While phytochemicals are also available in supplemental forms, there is limited evidence to suggest that they offer the same health benefits as those obtained through dietary intake.

These compounds, classified as secondary plant metabolites, exhibit various biological properties, including antioxidant activity, antimicrobial effects, modulation of detoxification enzymes, immune system stimulation, reduction of platelet aggregation, regulation of hormone metabolism, and potential anticancer effects. While over a thousand phytochemicals have been identified, many remain unknown. Although plants primarily produce these compounds for self-protection, recent research has shown that numerous phytochemicals also play a significant role in safeguarding human health and preventing disease [13,14].

Key Classes of Bioactive Phytochemicals

The term "phytochemicals" encompasses a diverse group of bioactive compounds found in nature, including steroids, flavonoids, polyphenols, and saponins. These substances are known to exert significant physiological effects on human health [15].

Flavonoids

To date, over 4,000 flavonoids of plant origin have been identified. They are predominantly found in green plants and primarily exist as glycosides in various plant parts, including leaves, flowers, stems, and roots. Structurally, flavonoids consist of two benzene rings, with major subclasses such as chalcones, flavones, flavanols, flavanones, anthocyanins, and isoflavones, many of which exhibit vibrant colors [16].

Phenylpropanoids

Phenylpropanoids are exclusively synthesized by plants and microorganisms through the shikimate pathway, which generates essential aromatic amino acids like phenylalanine and tyrosine. Since these amino acids are crucial for animals and humans but cannot be synthesized by them, plants serve as an essential dietary source within the food chain [16].

Terpenes

Terpenes represent a structurally diverse and extensive group of metabolites, with over 35,000 different compounds identified to date. Composed of isoprene units, they undergo cyclization reactions that contribute to their distinct structures. Based on the number of isoprene units in their carbon skeleton, terpenes are classified into various subgroups. Terpenoids, a modified form of terpenes, include bioactive metabolites with antitubercular, anticancer, anxiolytic, and mutagenic properties [17].

N-Containing Compounds

Alkaloids are cyclic organic compounds that contain nitrogen, though they are relatively rare in nature. They are generally soluble in aqueous solutions, allowing for efficient extraction through nitrogen protonation. This group includes well-known compounds such as caffeine, nicotine, cocaine, and morphine, which exhibit anxiolytic, analgesic, and hallucinogenic properties, often affecting the central nervous system. Despite being a small class of metabolites, alkaloids account for nearly 50% of plant-derived pharmaceuticals [18].

Techniques for Phytochemical Analysis

The separation of bioactive compounds from plant extracts remains challenging due to their varying polarities. To isolate these compounds, techniques such as TLC, HPTLC, paper chromatography, column chromatography, gas chromatography, OPLC, and HPLC are commonly used. Once purified, these compounds are analyzed for their structure and biological activity [19].

a) **Paper Chromatography**

Paper chromatography uses a sheet of filter paper as the inert solid phase for separation. It offers advantages such as simplicity, reliable reproducibility of Rf values, and dual functionality as both a support and a separation medium. In this technique, a sample is applied near the bottom of the filter paper, which is then placed in a chromatographic chamber with a solvent. The solvent moves upward by capillary action, carrying soluble molecules. The porosity of the paper affects the solvent movement, with low-porosity paper slowing the process and thicker paper increasing sample capacity [20].

b) **Thin Layer Chromatography (TLC)**

Thin layer chromatography (TLC), first practically applied by Stahl, offers advantages over paper chromatography, including versatility, speed, and sensitivity. It is an adsorption chromatography technique where compounds are separated based on their interaction with a thin layer of adsorbent on a plate. TLC is primarily used for separating low molecular weight compounds, with different adsorbents employed for various separations [21].

Table 1: Different adsorbent used to separate various compounds

Sr. No.	Adsorbent	Use to Separate
1	Silica gel	Amino acids, alkaloid, sugars, fatty acids, lipid etc.

2	Aluminium	Alkaloids, phenols, steroids, vitamins and carotenes.
3	Celite	Steroids and inorganic cations
4	Cellulose powder	Amino acids, food dyes, alkaloids
5	Starch	Amino acids
6	Sephadex	Amino acids, proteins

Methods of Detection

Fourier-transform infrared spectroscopy (FTIR)

Fourier-transform infrared (FTIR) spectroscopy is a powerful technique used to identify the functional groups present in plant extracts, aiding in the identification and structure determination of molecules. Samples for FTIR analysis can be prepared in several ways. For liquids, a drop of the sample is placed between two sodium chloride plates, forming a thin film. Solid samples are typically mixed with potassium bromide (KBr) and compressed into a thin pellet for analysis. Alternatively, solid samples can be dissolved in a solvent, such as methylene chloride, and a few drops of the solution are placed onto a High Attenuated Total Reflectance (HATR) plate. Spectra are then recorded in terms of percentage transmittance. Specific peaks at certain wave numbers are assigned to particular bonds and functional groups according to the reference provided in the Varian FTIR instrument manual [25,26].

MATERIAL & METHODS:

Some Pharmacognostic study of plant:

Ash Value:

Weigh and ignite flat, thin, porcelain dish or a tared silica crucible. Weigh about 2 g of the powdered drug into the dish/crucible. Support the dish on a pipe-clay triangle placed on a ring of retort stand. Heat with a burner, using a flame about 2 cm high and supporting the dish about 7 cm above the flame, heat till vapours almost cease to be evolved, then lower the dish and heat more strongly until all the carbon is burnt off. Cool in a desiccator. Weigh the ash and calculate the percentage of total ash with reference to the air dried sample of the crude drug.

Calculations:

Weight of the empty dish = x

Weight of the drug taken = y

Weight of the dish + Ash (after complete incineration) = z

wt. of the ash = (z-x) g

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'y' g of the crude drug gives (z-x) g of the ash

100 g of the crude drug gives 100 /y x (z-x) g of the ash

Total ash value of the sample = $100 (z-x) / y$

e.g. Total ash value of Cinchona bark is not more than 4%.

Extractive Value:

- Useful for the evaluation of a crude drug.
- Give idea about the nature of the chemical constituents present in a crude drug.
- Useful for the estimation of constituents extracted with the solvent used for extraction.
- Employed for material for which as yet no suitable chemical or biological assay exists.

1. Alcohol Soluble extractives

Procedure:

- Weigh about 4 g of the coarsely powdered drug in a weighing bottle and transfer it to a dry 250 ml conical flask.
- Fill a 100 ml graduated flask to the delivery mark with the solvent (90% alcohol). Wash out the weighing bottle and pour the washings, together with the remainder of the solvent into the conical flask.
- Cork the flask and set aside for 24 hours, shaking frequently. (Maceration).
- Filter into a 50 ml cylinder. When sufficient filtrate has collected, transfer 25 ml. of the filtrate to a weighed, thin porcelain dish, as used for the ash values determinations.
- Evaporate to dryness on a water-bath and complete the drying in an oven at 105° C for 6 hrs.
- Cool in a desiccator for 30 minutes and weigh immediately.

Calculation:

25 ml of alcoholic extract gives = x g of residue

100 ml of alcoholic extract gives = 4x g of residue

since ... 5 g of air dried drug gives 4x g of alcohol (90%) soluble residue.

∴ 100 g of air dried drug gives 80x g of the alcohol (90%) solute residue.

Alcohol (90%) soluble extractive value of the sample = 80x%

Water Soluble Extractives:

Procedure:

Steps are similar to those mentioned in the previous experiment.

Use chloroform water instead of alcohol (Chloroform acts as a preservative).

Biological study of Plants:

Brine Shrimp Lethality Assay:

Brine shrimp (*Artemia salina*) eggs were hatched in artificial sea water prepared from commercial sea salt 38 g/L. A lamp was placed above the open side of the tank to attract the hatched shrimps close to the tank wall. After 24 hours, the shrimps matured as nauplii (*Artemia salina*) and were ready for the assay. The brine shrimp lethality bioassay was carried out on the extract using the standard procedure. Twenty milligrams of the extract were dissolved in 1 ml ethanol to give a crude extract concentration of 20 mg/mL. A two-fold serial dilution was carried out with salt water to obtain a test solution in the range of 0.1 – 10 mg/mL. Each concentration was tested in triplicate. A test-tube containing in 5 ml of salt water was used as the negative control. To obtain test concentrations ranging from 0.02 to 30 mg/ml. A suspension of larvae (0.1 ml), containing about 10 – 15 larvae, was added into each test tube and incubated for 24 hours. The test tubes were then examined, and the number of dead larvae in each bottle was counted after 6, 12, and 24 hours. The total number of shrimps in each bottle was counted and recorded. The death percentage and lethal concentration (LC50) were determined using statistical analysis).



Anti-inflammatory Activity

The anti-inflammatory activity of various extracts of leaves of *Artemisia Japonica*, *Martynia annua*, *Erythrina variegata*, *Jatropha curca* was evaluated using the in vitro HRBC (Human Red Blood Cell) membrane stabilization method. The experiment focused on assessing the membrane protection potential of the methanolic extract of *Artemisia Japonica* at different concentrations.

1. Materials

- Fresh human blood (collected from healthy volunteers)
- Alsever's solution:
- Methanolic extract of *Gardenia coronaria*
- Spectrophotometer (set at 560 nm)

2. Procedure

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1. Preparation of HRBC Suspension:

- Blood was collected from healthy volunteers and mixed with an equal volume of Alsever's solution.
- The mixture was centrifuged, and the pellet (HRBC) was washed with isosaline.
- 1 mL of the HRBC suspension was prepared for further analysis.

2. Treatment Groups:

- Equal volumes of the HRBC suspension were mixed with test solutions at concentrations of 10,20,40,60,80,100µg/mL.
- A control group contained Alsever's solution with blood but no drug.

3. Incubation and Measurement:

- All assay mixtures were incubated at 37°C for 30 minutes and centrifuged.
- The supernatant's hemoglobin content was measured using a spectrophotometer at 560 nm.

Result and Discussion:

As we performed ash value study of plant extract we got a result as mention in Table 2. It shows a greater Total ash value for the sample 2 while lowest is of sample 1. While acid insoluble ash for sample 3 is more and sample 1 is less. But water soluble ash for the sample 1 is highest and for sample 3 is lowest.

Table 2: Ash value of plant extracts

Sample	Ash Value		
	Total Ash	Acid Insoluble	Water Soluble
Sample 1	7.5%	8.0%	7.5%
Sample 2	30.5%	10.0%	0.5%
Sample 3	18.5%	12.0%	0.1%
Sample 4	16.5%	11.5%	1.0%

Extractive values:

Extractive value indicates the approximate amount of active phytoconstituents present in plant material when extracted with a particular solvents. It also gives an idea about the nature (polar or non polar) of the chemical constituents present in the crude drug.

In present study the extractive values of sample of *Artemisia Japonica*, *Martynia annua*, *Erythrina variegata*, *Jatropha curca* was determine using ethanol, water & Petroleum ether as a solvent.

Table 3: Ash value of plant extracts

Sample	Extractive Value

	Ethanollic extract	Water extract	Petroleum Ether Soluble
Sample 1	1.6%	6.5%	7.5%
Sample 2	6.0%	1.0%	0.5%
Sample 3	11%	2.75%	0.1%
Sample 4	14.5%	14.25%	1.0%

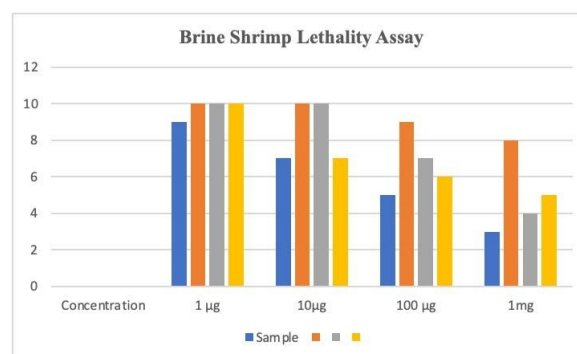
Brine Shrimp Lethality Assay:

The data show the response of four plant samples at different concentration (1 µg, 10 µg, 100 µg, and 1mg). The values indicate the biological activity (such as zone of inhibition or inhibitory response).

FORMULA:-Percentage of Death (%): (Total nauplii Alive nauplii) X (100%/Total nauplii)

Table 4: Brine shrimp lethality assay of plant extracts

Concentration	Sample			
	Sample 1	Sample 2	Sample 3	Sample 4
1 µg	09	10	10	10
10µg	07	10	10	07
100 µg	05	09	07	06
1mg	03	08	04	05



Anti-inflammatory report:

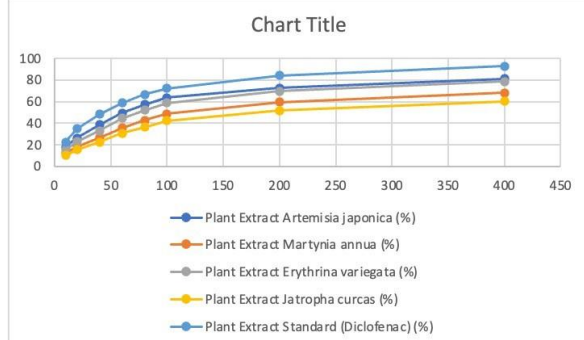
The percentage of membrane protection at different concentrations of the methanolic extract of *Artemisia Japonica*, *Martynia annua*, *Erythrina variegata*,

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Jatropha curca was calculated. The results are summarized in the table and visualized in the graph below

Table 5: Anti-inflammatory studies of plant[8] extracts

Concentration (µg/mL)	Artemisia japonica (%)	Martynia annua (%)	Erythrina variegata (%)	Jatropha curcas (%)	Standard (Diclofenac) (%)
10	18.2	12.5	15.4	10.2	22.6
20	26.4	18.3	22.8	15.6	34.8
40	38.7	26.5	33.2	22.4	48.3
60	49.5	35.8	44.6	30.7	58.9
80	57.3	42.6	52.1	36.4	66.5
100	63.8	48.9	58.7	42.3	72.4
200	72.6	59.4	69.5	51.8	84.2
400	81.4	68.2	78.6	60.3	92.7



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