

Comparative Qualitative and Spectroscopic Evaluation of Phytochemicals in Medicinal Plants

A Thesis Submitted

in Partial Fulfillment of the Requirements for the Degree of

MASTER OF SCIENCE IN BIOTECHNOLOGY

in

BIOTECHNOLOGY

by

CHETNA RANI & AKANSHA

(Roll No. 24/MSCBIO/53)

(Roll No. 24/MSCBIO/26)

Under the Supervision of

DR. NAVNEETA BHARADVAJA

**Associate Professor, Department of Biotechnology
Delhi Technological University**



To the

Department of Biotechnology

DELHI TECHNOLOGICAL UNIVERSITY

(Formerly Delhi College of Engineering)

Shahbad Daultpur, Main Bawana Road, Delhi-110042

May, 2026

ACKNOWLEDGEMENT

We would like to express our sincere gratitude to my supervisor, **Dr. Navneeta Bharadvaja**, for providing us the opportunity to carry out this research work and for her continuous guidance, encouragement, and valuable suggestions throughout the study. Her knowledge, motivation, and constant support inspired us to complete this work successfully. We are deeply thankful for her insightful feedback and encouragement, which helped us improve our understanding and research skills. We would also like to express our heartfelt thanks to the Department of Biotechnology, Delhi Technological University, for providing the necessary facilities and support during the completion of this project.

We are highly thankful to **Ms. Harshita Singh (Ph.D. Scholar)** for her valuable guidance, support, and encouragement throughout the research work. Her suggestions and assistance greatly helped us during the project.

We would also like to sincerely thank Mr. Chhail Bihari and Mr. Jitendra Sir for their constant support and cooperation during the completion of this work. We extend our deepest gratitude to our parents and family members for their unconditional love, prayers, motivation, and support, which encouraged us throughout our academic journey. Finally, we would like to thank all those who directly or indirectly contributed to the successful completion of this research work.

Chetna Rani & Akansha



DELHI TECHNOLOGICAL UNIVERSITY

(Formerly Delhi College of Engineering)

Shahbad Daultapur, Main Bawana Road, Delhi-42

CANDIDATE'S DECLARATION

WE, **CHETNA RANI and AKANSHA** Roll No. **2K24/MSCBIO/53, 2K24/MSCBIO/26** hereby declare that the dissertation entitled “**Comparative Qualitative and Spectroscopic Evaluation of Phytochemicals in Medicinal Plants**” submitted to the Department of Biotechnology, Delhi Technological University in partial fulfilment of the requirements for the award of the degree of **Master of Science**, is a bona fide record of the original research work carried out by us during the period from **January 2026 to May 2026** under the supervision of **Dr. Navneeta Bharadvaja**.

We further declare that the work presented in this thesis has not been submitted, either in part or in full, for the award of any other degree, diploma, or fellowship of this or any other university/institute.

1. Our conference paper was presented and accepted at an **SFE-indexed international conference** with the following details:

Title of the paper: Role of *Bacopa monnieri* and Bacosides in modulating Alzheimer disease pathophysiology

Name of Authors: Chetna Rani, Akansha, Dr. Navneeta Bharadvaja

Conference name: International Conference on Biomedical Technologies and Bioinformatics (ICBTBI)

Conference Indexing: Scopus/Crossref/Google Scholar/SFE

Status of paper: Presented

Accepted Date of paper acceptance: May 8th, 2026

Place: Hyderabad, India



DELHI TECHNOLOGICAL UNIVERSITY

(Formerly Delhi College of Engineering)

Shahbad Daultapur, Main Bawana Road, Delhi-42

CERTIFICATE BY THE SUPERVISOR

It is certified that **CHETNA RANI and AKANSHA, Roll No. (24/MSCBIO/26) and (24/MSCBIO/53)** have carried out their research work presented in this thesis entitled “**Comparative Qualitative and Spectroscopic Evaluation of Phytochemicals in Medicinal Plants**” for the award of Degree of Masters of Science in Biotechnology and submitted to the Department of Biotechnology, Delhi Technological University, Delhi under my supervision. The thesis embodies results of original work, and studies are carried out by the student themselves, and the contents of the thesis do not form the basis for the award of any other degree to the candidate or to anybody else from this or any other University/Institution.

Prof. Yasha Hasija

Head of Department

Department of Biotechnology

Delhi Technological University

Dr. Navneeta Bharadvaja

Associate Professor

Department of Biotechnology

Delhi Technological University

Date:

Comparative Qualitative and Spectroscopic Evaluation of Phytochemicals in Medicinal Plants

CHEटना RANI & AKANSHA

Delhi Technological University, Delhi, India

E-mail: chetnarani2000@gmail.com & akanshakherpa@gmail.com

ABSTRACT

The objective of this research was to analyze the phytochemical constituents and medicinal values of certain medicinal plants through qualitative, quantitative, and spectroscopic analysis. The aqueous extracts of selected medicinal plants that is *Euphorbia milii*, *Matricaria chamomilla*, *Parthenium hysterophorus* and *Stevia rebaudiana*, were obtained, and certain phytochemical constituents were determined qualitatively. Qualitative analysis indicated the existence of alkaloids, phenols, tannins, flavonoids, saponins, terpenes, sterols, and quinones in the plant extracts. Quantitative analysis emphasized variations in the concentration of phytochemicals in the selected plants. Further, UV–Visible spectroscopy was performed for identification and estimation of phytochemicals. FTIR spectroscopy was performed to determine functional groups in bioactive compounds. The results emphasize the significance of medicinal plants as natural sources of phytochemicals with antioxidative and therapeutic effects.

LIST OF FIGURES

Figure No.	Title	Page No.
Fig. 1	Steps for the Extraction of Phytochemicals	27
Fig. 2	UV-Vis Graph Absorbance for Phenol	43
Fig. 3	FTIR Graph 1	44
Fig. 4	FTIR Graph 2	45
Fig. 5	FTIR Graph 3	46
Fig. 6	FTIR Graph 4	47

LIST OF TABLES

Table No.	Title	Page No.
Table 1	Qualitative Test for Different Phytochemicals	31
Table 2	Phytochemical Analysis of <i>Euphorbia milli</i>	32
Table 3	Phytochemical Analysis of <i>Matricaria chamomilla</i>	33
Table 4	Phytochemical Analysis of <i>Parthenium hysterophorus</i>	35
Table 5	Phytochemical Analysis of <i>Stevia rebaudiana</i>	36
Table 6	Comparative Absorbance Analysis at 765 nm	41
Table 7	Interpretation of Possible Compound in Chamomile by FTIR Graph	42
Table 8	Interpretation of Possible Compound in <i>Euphorbia milli</i> by FTIR Graph	43
Table 9	Interpretation of Possible Compound in <i>Parthenium hysterophorus</i> by FTIR Graph	44
Table 10	Interpretation of Possible Compound in <i>Stevia rebaudiana</i> by FTIR Graph	45

TABLE OF CONTENTS

CONTENTS	
Acknowledgement	2
Candidate's Declaration	3
Certificate by Supervisor	4
Abstract	5
List of Figures	6
List of Tables	7
CHAPTER I: INTRODUCTION	10
1.1 GENERAL INTRODUCTION	11
1.2 RELEVANCE OF THE PROJECT	12
1.3 OBJECTIVE OF THE STUDY	12
1.4 MEDICINAL PLANTS SELECTED FOR STUDY	12
CHAPTER II: REVIEW OF LITERATURE	12
2.1 <i>Euphorbia milli</i>	12
2.2 <i>Matricaria chamomilla L</i>	14

2.3 <i>Parthenium hysterophorus L</i>	16
2.4 <i>Stevia rebaudiana</i>	18
2.5 Introduction to Phytochemicals	19
2.6 Extraction and Isolation of Phytochemicals	24
2.7 Phytochemicals screening techniques	25
2.8 UV- Vis Spectroscopy	25
2.9 FTIR Spectroscopy	26
CHAPTER III: METHODOLOGY	27
3.1 Extraction of Sun Dried Leaves	27
3.2 Extraction of Fresh Leaves	28
3.3 Qualitative Estimation of secondary metabolites	29
3.4 Protocol for Qualitative Phytochemical Screening	30
3.5 Quantitative Estimation of Total Phenolic Content by UV-VIS Spectrophotometry	32
3.6 Quantitative Estimation of Alkaloid Amount by UV-VIS Spectrophotometry	33
CHAPTER IV : OBSERVATIONS AND RESULTS	34
4.1 Qualitative Analysis of Aqueous solution of dry and fresh extracts	34
4.2 Quantitative Analysis	43
a. UV- Vis Spectrophotometer Analysis for Phenolic Content	43
b. FTIR Analysis of selected medicinal plants	44
CHAPTER V : DISCUSSION	48
REFERENCES	

CHAPTER 1:- INTRODUCTION

Phytochemicals are natural derived compounds occurring in medicinal plants, specifically in plant parts like leaves, roots, stems, bark, flowers, seeds, etc., having a specific mechanism and protective effects.[1][2] Numerous bioactive chemicals in plants affect different physiological functions in humans; hence, medicinal properties in plants can be attributed mainly to phytochemicals.[3] Metabolites in plants are divided mainly into two categories: primary metabolites and secondary metabolites. Primary metabolites, such as sugars, amino acids, lipids, proteins, and chlorophyll, are vital in normal growth and development processes, while SMs include alkaloids, flavonoids, tannins, terpenoids, saponins, phenols, essential oils, and cardiac glycosides.[4]

These secondary metabolites display various biological functions such as antioxidant, antiapoptotic, antiaging, anticarcinogenic, anti-inflammatory, antiatherosclerosis, cardiovascular protecting, antiangiogenic, and antiproliferative properties.[5] As a result of their medicinal properties, phytochemical screening techniques have been adopted widely as a means of detecting and identifying bioactive compounds in plants, which could also form the foundation for drug development in the future. Phytochemical qualitative screening can be done to determine different kinds of biologically active compound.[6]

Secondary metabolites that play an important role in defending themselves against pathogens, herbivorous attack .[7] These adverse environmental conditions.[8] Phenolics, alkaloids, terpenes, tannins, and saponins are common secondary metabolites found in plants with important pharmacological applications.[8] Plants with significant amounts of secondary metabolites are important sources of natural medicinal substances and can be used in the development of drugs.[9] Alkaloids are nitrogenous compounds known for their numerous applications in modern medicine due to their high therapeutic potential.[7] These nitrogenous metabolites are important for antimicrobial, analgesic, and anti-inflammatory actions.[8] Some of the phytochemicals including alkaloids and polyphenols have proved effective in providing antimicrobial, anticancer, and anti-inflammatory actions.[7] Phenolics are important secondary metabolites found in plants that have been shown to have significant antioxidant capacity through free-radical scavenging.[10]

Phenolic metabolites provide important contributions towards the antioxidant potential due to their hydrogen donating action against harmful free radicals.[8] Polyphenols are important phytochemicals known for their antioxidant capacity due to their ability to protect against oxidation.[7] Flavonoids and phenolic components are important metabolites in protecting against free radicals by neutralizing their harmful effects. Therapeutic importance of phenolic compounds can be associated to their ability to protect against oxidative stress-related diseases. [10] Tannins are polyphenolic phytochemicals known for their wide-spectrum antibacterial activity against pathogenic microorganisms. Antibacterial properties of tannins can be attributed to inhibition of enzymes, interference with cellular processes, and prevention of nutrient transport.[11]

Natural antioxidants in the form of polyphenols from plants have drawn substantial attention in modern medicine.[11] Saponins are glycosides occurring in medicinal plants. Antioxidant activity of saponins from medicinal plants has been attributed to their radical scavenging and membrane protection activity.[12]

Euphorbia milii is a valuable medicinal plant whose use for skin inflammation treatment, because of the presence of diterpenoids, triterpenoids, flavonoids, and macrocyclic lactones. Some biological activities observed in the extracts obtained from *Euphorbia milii* include antioxidative, antimicrobial, anti-inflammatory, anticancer, antiparasitic, and other health benefits.[14] *Matricaria chamomilla* one of the earliest used medicinal plants in treating inflammatory diseases, gastrointestinal issues, and skin infections. Bioactive components found in chamomile include flavonoids, terpenoids, coumarins, and essential oils, giving this medicinal plant excellent antioxidative, anti-inflammatory, antimicrobial, wound-healing, and mild sedative activities.[15]

Parthenium hysterophorus, having important phenolic compounds, flavonoids, and sesquiterpene lactones, contributing to the pharmacological significance of this medicinal plant. *Parthenium hysterophorus* has shown excellent antioxidative and anti-inflammatory activities; traditionally, this plant was used to treat skin problems, fever, inflammations, and infectious diseases.[16] *Stevia rebaudiana* is a popular because of the presence of intensely sweet steviol glycosides along with stevioside, rebaudioside, flavonoids, and phenols.[17]

Traditionally, *Stevia rebaudiana* was used for managing diabetes, high blood pressure, and metabolic problems; antioxidants and other pharmacological properties of this medicinal plant are due to the phenols and natural glycosides, which are potent free radical scavengers.[13], [18] The process of extraction and spectroscopic characterization of phytochemicals through analytical methods such as UV–Visible and FTIR spectroscopy .[13] Because of their antimicrobial, anticancer, anti-inflammatory, and immunostimulant effects, saponins have received importance in medicinal research.[12]

One popular technique in the studies of medicinal plants is aqueous extraction, which is safe, cheap, and eco-friendly concerning the extraction of polar active phytochemicals.[19] Qualitative phytochemical profiling is done to check the existence of certain secondary metabolites in medicinal plants, namely, alkaloids, phenols, tannins, and saponins. The quantification of phytochemicals is an important procedure in determining the content of medicinal substances in plants. UV-Vis spectroscopy is a fast and efficient method for the identification of phytochemicals.[20] This technique is helpful for both quantitative and qualitative analysis on the basis of absorption spectra , indicates presence of phenolics, flavonoids, and other antioxidants in plant extracts.[21], [22]

FTIR spectroscopy is an indispensable method for the determination of functional groups and the analysis of the structure of biologically active phenolics and flavonoids.[13], [22] Phytochemical evaluation and spectroscopic characterization can provide scientific substantiation for the pharmacological properties of medicinal plants.[20]

1.2 Relevance of the project

Bioactive secondary metabolites are regarded as important sources with various pharmaceutical properties. Alkaloids, phenolic, tannins, and saponins are among the phytochemical compounds that possess antioxidant, antimicrobial, anti-inflammatory and medicinal properties. Natural plant-based products have gained immense popularity in recent times as alternative treatment methods, since they pose less risk compared to synthetic drugs. UV-Visible and FTIR spectroscopy are commonly used in identifying plant extracts through phytochemical and functional group analyses.

1.3 Objective of the study

1. Conduct qualitative phytochemical analysis for detection of phytochemical constituents.
2. Phytochemical analysis of plant extracts using UV-Visible Spectroscopy.
3. Determination of functional groups of plant extracts using FTIR spectroscopy.

The current study may provide scientific information regarding phytochemistry of medicinal plants and their medicinal relevance. This could also be useful in promoting phytochemicals of plant origin in future pharmaceutical applications.

1.4 Medicinal Plants Selected for the Study

1. *Euphorbia milli*
2. *Matricaria chamomilla*
3. *Parthenium hysterophorus*
4. *Stevia rebaudiana*

Chapter 2: Review of Literature

Medicinal plants have gained considerable scientific interest in recent years because their phytochemical composition is rich, and also because they have a broad range of therapeutic uses. From these plants, secondary metabolites appear to have major pharmacological activities like antioxidant effects, antimicrobial properties, anti-inflammatory actions and even anticancer activities. In the next section some medicinally relevant plants are mentioned that have been studied for their phytochemical potential in general terms.

2.1 *Euphorbia milli*

2.1.1 Botanical Description

Euphorbia milii is a scandent succulent, ornamental shrub because its thick fleshy brownish stems carry sharp spines.[23] It has alternate obovate leaves, and it forms brightly coloured bracts around flowers that are relatively inconspicuous. [24] Native to Madagascar, *E. milii* is well adapted for dry and semi dry conditions, and it contains toxic milky latex in every part of the plant and produces triangular-ovoid capsules with greyish brown seeds, and it flower throughout the year.[23]

2.1.2 Phytochemical Constituents

Phytochemical investigations of *Euphorbia milii* have showed that diverse secondary metabolites are present like triterpenoids, flavonoids, alkaloids, diterpenoid and saponins which are notable for medicinal value [24][23]In fact, among these constituents the diterpenoids class seems to be the dominant group of phytochemicals and it is linked with different biological actions of the plant [14][25] UHPLC-MS profiling along with phytochemical screening studies, also supported that coumarins, cardiac glycosides and terpenoid compounds can be found in various extracts of *E. milii*. [24] [26].

2.1.3 Medicinal Importance

Euphorbia milii possesses significant medicinal value because it contains a diverse phytochemical composition and also exhibits diverse pharmacological action [24] In tradition medicine, the plant has often been used for skin infections, inflammatory problems, and wound related conditions with people relying on it as part of home remedies [28] Many studies report that extracts and latex from *E. milii* can act as antioxidants, as well as antimicrobials and anti-inflammatory agents, mostly because it contains various secondary metabolites [24], [28]The overall pharmacological impact is often linked with tannins, flavonoids, saponins, and phenolic constituents which are connected with tissue repair and therapeutic potential [25]

2.1.4 Anti-Microbial Activity

In antimicrobial assays it came out that *E. milii* extracts exhibited inhibitory action against *Staphylococcus aureus*, also *Escherichia coli*, and several fungal pathogens[24], [27] The plant's antibacterial and antifungal properties is because of the presence of flavonoids, tannins and other phytochemical components [27]

2.1.5 Anti-Oxidant Activity

The methanolic and floral extracts from *E. milii* exhibited significant DPPH radical scavenging action, which points to a strong antioxidant potential [23], [29], [30]. The total phenolic content was very high, together with the flavonoids and saponins, these constituents significantly contributed to explain the antioxidant and the cytotoxic characteristics of the plant extracts [31]. Also, the antioxidant effects of *E. milii* have been associated with phenolic compounds, these exhibit free radical scavenging, and they can participate in metal ion binding too, thereby supporting the observed antioxidant potential [30]. Additionally, the phenolic compounds and flavonoids found in *Euphorbia* species play a major role in anti-inflammatory mechanisms, mostly via free radical scavenging activity [25].

2.1.6 Toxicological and Phytochemical Investigations

The milky latex of *Euphorbia milii* has diterpenoid constituents with both pharmacological and irritant traits and upon excessive exposure it can lead to skin irritation and inflammatory reactions, due to the presence of toxic compounds [14], [25]. Even though it is toxic at higher doses, *E. milii* still shows a noticeable medicinal value, partly due to its diverse phytochemical makeup and therapeutic usefulness [28]. Overall, these phytochemical screening and extraction reports highlight that *Euphorbia milii* is scientifically significant as a medicinal plant with both Therapeutic effects, and toxicological relevance [26], [27], [28], [32].

2.2 *Matricaria chamomilla* L.

2.2.1 Introduction and botanical description

Matricaria chamomilla L. often referred to as German chamomile and spread broadly across Europe and Western Asia. [33] Out of all the parts, the flowers are usually treated as the main medicinal part, and they're known for being particularly rich in essential oils as well as in other bioactive phytochemical components. [15] Because it contains pharmacologically active constituents, chamomile gets used in traditional medicine, in cosmetic products and in the more general herbal market. [33]

2.2.2 Phytochemical Constituents

Major bioactive constituents that have been reported in chamomile include apigenin, quercetin, luteolin, and chamazulene, and they seem to really help explain a lot of the plant's medicinal, plus pharmacological properties. [34] The essential oil from chamomile is especially abundant in sesquiterpenes and other terpenoid compounds like chamazulene, which are linked with anti-inflammatory effects and a kind of gentle calming activity. [33] Several studies that used

methanolic and ethanolic extracts of *M. chamomilla* have shown a high phytochemical level, along with notable biological activity, mostly because phenols and flavonoids are extracted very efficiently.[34]

2.2.3 Anti-Inflammatory and Sedative Activity

Matricaria chamomilla has been widely described as having strong anti-inflammatory effects, and it's mostly linked to flavonoids along with terpenoid related constituents that are found in the flower extracts and essential oils [33]. In other words, those components show up in the extracts, not just one part. Some bioactive molecules like chamazulene are reported to hinder inflammatory signals and also help reduce swelling in tissues, so the overall therapeutic impact of chamomile preparations becomes more reliable, and also more consistent [15]. Chamomile contains sedative properties and in traditional usage it has been used for anxiety, nervousness and even sleep related troubles [33]. Not only that, people often describe it as a calming herb even if the reasons are biochemical. One particular flavonoid, apigenin, that is found in chamomile has been said to affect benzodiazepine receptors within the brain, which can lead to anxiolytic and sleep-inducing outcomes [35]. A number of pharmacological investigations have also shown that chamomile extracts can act as an antispasmodic agent and a muscle relaxing remedy, which may be useful when dealing with gastrointestinal spasms.[15]

2.2.4 Gastroprotective Uses

Matricaria chamomilla has been used for a long time to help with different gastro intestinal problems, like indigestion, gastric irritation, and intestinal spasms, mainly because it works in an antispasmodic way and it feels calming.[15] A number of pharmacological investigations have described gastroprotective and anti-ulcer effects from chamomile extracts. This could be linked with blocking inflammatory signaling and lowering the inflammation on the gastric lining.[35] Chamomile products are often chosen for reducing belly discomfort and digestive spasms since their flavonoids and essential oil components show muscle relaxing actions.[15]

2.2.5 Phytochemical and Spectroscopic Studies

UV-Visible spectrophotometric work was used to detect high phenolic and flavonoid levels. FTIR spectroscopic characterization of chamomile extracts indicated functional groups such as hydroxyl and carbonyl group and this gives a spectral fingerprinting effect and also instrumental validation of those pharmacologically relevant phytochemicals.[36]

2.2.6 Conclusion

Matricaria chamomilla is often treated as a major medicinal herb, mainly because it has a long traditional background, and also because it shows a wide range of pharmacological capabilities that have been confirmed through scientific studies. [33] The secondary metabolites help explain why it is therapeutically valuable for pharmaceutical, cosmetic, and herbal applications.[34] In addition, phytochemical screening, together with UV-Visible and FTIR spectroscopic examinations, has given useful information about how the biologically active constituents in chamomile extracts can be characterized and also validated.[36]

2.3 *Parthenium hysterophorus* L.

2.3.1 Introduction and botanical description

Parthenium hysterophorus L. is an annual herbaceous weed, usually called as carrot grass, congress grass and bitter weed. Morphologically, *P. hysterophorus* shows an erect branched stem, leaves that are finely divided, small white flowers, and well-developed root system, which helps it to survive under wide range of environmental conditions. Even though it is generally treated as a noxious invasive weed, several segments of the plant—leaves, flowers, and also whole plant extracts—have slowly gained attention for phytochemical and pharmacological studies.[38]

2.3.2 Phytochemical Constituents

Phytochemical investigations of *Parthenium hysterophorus*, have shown a range of secondary metabolites, like alkaloids, phenolic substances, tannins, saponin, flavonoids, terpenoids, and glycosides.[38] This plant seems especially abundant in sesquiterpene lactones such as parthenin, hysterin, and ambrosin, and these are widely viewed as central biologically active components that link with a number of pharmacological as well as toxicological activities.[39] When researchers used methanolic as well as ethanolic extracts of *P. hysterophorus*, the results showing strong phytochemical richness, suggesting that polar solvents work quite well for extract out phenols, flavonoids and various other compounds that are useful in medicine. [40] Organ specific phytochemical profiling has also indicated that chlorogenic acid, caffeic acid, and other phenolic constituents are not uniformly spread.[43]

2.3.3 Antimicrobial Activity

In various in vitro studies, methanolic and ethanolic extracts of *Parthenium hysterophorus* have shown antibacterial, as well as antifungal, effects against different pathogenic microorganisms. [38] For leaf extracts and flower extracts, the antimicrobial potential seems to be mostly linked with phenolic substances, tannins, flavonoids, and terpenoid related constituents, although the exact contribution may vary between reports.[39] Many of the phytochemicals found in *P. hysterophorus* are described to slow down microbial development via actions like cell membrane disruption, protein denaturation, and also by disturbing essential metabolic pathways. [38] Because it contains biologically active secondary metabolites, *Parthenium hysterophorus* has drawn interest as a plausible reservoir of medicinal compounds, with antimicrobial value.[39]

2.3.4 Antioxidant Activity

Parthenium hysterophorus is shown to have antioxidant phytochemicals, mostly phenolic compounds along with flavonoids, and this is linked with its ability for free-radical scavenging.[40] Studies that used DPPH together with FRAP assays suggest that the

methanolic extract of *P. hysterophorus* shows antioxidant activity, as well as ability for lowering oxidative stress. [41] A number of investigations also found a favourable relationship between total phenolic content and antioxidant performance.

2.3.5 Allergy and toxicological effects

Parthenium hysterophorus is very often treated as a harmful invasive weed, and it can cause uncomfortable consequences in humans and animals too. This occurs because the plant carries allergenic compounds along with other biologically active phytochemicals, so it can set off toxic reactions. [37]

Also, plant extracts of *Parthenium hysterophorus* have been shown to cause cytotoxic and irritant effects. That suggests they might provoke inflammatory and oxidative stress related cellular harm. Even with that toxicological weight and the allergenic profile, researchers still keep looking at it pharmacologically. The reason is its varied bioactive constituents and the possible medicinal uses. [40]

2.3.6 Phytochemical and Spectroscopic Studies

From qualitative phytochemical screening of *Parthenium hysterophorus* extracts, it was found that there are alkaloids, phenols, tannins, saponins, along with other biologically active secondary metabolites in several parts of the plant. [42] Then quantitative phytochemical assessments, and spectrophotometric investigations, showed a clear distinction in phenolic as well as flavonoid levels across leaves, flowers and other organs of *P. hysterophorus*. [41] In addition, UV–Visible spectrophotometric analysis has been used extensively, not only for estimation but also for characterization of phenolic compounds within methanolic and ethanolic extracts of the plant. [42] Moreover, FTIR characterization work indicated key functional moieties like O–H, C=O, C–O, and N–H, so the results enabled spectral fingerprinting and also acted as an instrumental validation for the phytochemicals that are present in *Parthenium hysterophorus* extracts. [41] Overall, phytochemical and spectroscopic observations have offered meaningful scientific support, about the medicinal relevance and the biological effects linked to the bioactive constituents in *Parthenium hysterophorus*. [42]

2.3.7 Conclusion

Parthenium hysterophorus is considered a phytochemically significant invasive weed, mainly because it contains a range of biologically active secondary metabolites with pharmacological and toxicological impacts. [37] This plant holds multiple medicinally relevant constituents, including phenols, flavonoids, and sesquiterpene lactones, and these molecules help drive its antimicrobial, antioxidant, anti-inflammatory, and other types of biological effects.

Even if it is slightly allergenic and toxic, *Parthenium hysterophorus* still attracts substantial scientific attention, partly due to its possible therapeutic uses and its pharmacological relevance. Phytochemical exploration together with spectroscopic characterization approaches like UV–Visible and FTIR analyses, are useful for detecting, describing, and confirming bioactive components present in *Parthenium hysterophorus*. Overall, the scientific references that are available indicate that *Parthenium hysterophorus* work as a promising source of

biologically active phytochemical substances, for upcoming pharmaceutical and medicinal investigations.[39]

2.4 *Stevia rebaudiana*

2.4.1 Introduction and Botanical Description

Stevia rebaudiana Bertoni, which is part of the family Asteraceae, is usually called stevia, sweet leaf, and sugar leaf. It is used as a natural low-calorie sweetener.[44] *S. rebaudiana* is a perennial herbaceous plant. It has a branched stem, leaves that are sessile and oppositely arranged and small white flowers. The root system is well developed, and it seems adjusted for warm climatic conditions.[45] In most phytochemical and pharmacological studies, the leaves and leaf extracts are the plant parts most often used. The main reason is that they contain high levels of sweet diterpene glycosides, along with other bioactive metabolites.[43] Because it supports nutraceutical, pharmaceutical, and food industry uses, *Stevia rebaudiana* has attracted a lot of scientific interest. It is identified as an economically important medicinal plant, with potential therapeutic effects.[45]

2.4.2 Phytochemical Constituents

Stevia rebaudiana has a broad set of biologically active secondary metabolites like phenolic compounds, flavonoids, tannins, saponins, alkaloids, terpenoids, and glycosides, and these together support its medicinal role, [45]. In particular, the plant is rich in sweet diterpene glycosides, such as stevioside, rebaudioside A, dulcoside, and steviol, which are often treated as the key bioactive components behind both the sweetness effect, and its therapeutic benefits. [46] Some phytochemical research also described chlorogenic acid and several other phenolic constituents in stevia, because these are linked with profound antioxidant potential and pharmacological effects in stevia leaf extracts. [47] Generally, the occurrence and level of phytochemicals tend to be higher in the leaves so, that part becomes the main focus for phytochemical and pharmacological studies. [45] Many publications have shown that the medicinal actions of *Stevia rebaudiana*, such as antidiabetic, antioxidant and anti-inflammatory effects are closely associated to its varied phytochemical composition [47]

2.4.3 Antidiabetic, and Hypoglycemic Activity

Stevia rebaudiana has shown some real antidiabetic potential, mostly because it can reduce blood glucose and also help in glucose metabolism, suggested by variety of experimental and clinical studies [48] The hypoglycemic effect of stevia is mainly linked with steviol glycosides like stevioside and rebaudioside A, and these are considered for diabetes treatment and management.[47] Several investigations have also claimed that stevioside can act in an insulinotropic way, hence it boosts insulin release from pancreatic β -cells that supports glycemic control. [45] People are consuming stevia as a natural low-calorie sugar alternative, not just out of preference, but because it gives sweetness without raising blood glucose.[48] Overall, pharmacological findings indicates that the antidiabetic activity of *S. rebaudiana* control the insulin signaling routes, better glucose usage, and also lowering the oxidative stress that commonly leads to hyperglycemia.[47]

2.4.4 Therapeutic and Pharmacological Importance

Stevia rebaudiana is known for strong antihypertensive activity, and supports cardiovascular health via vasodilatory actions and a more consistent reduction in blood pressure. [43] A number of investigations also indicate anti-inflammatory and antimicrobial effects of stevia extracts, and this is mostly connected with the presence of steviol glycosides, flavonoids, and phenolic compounds. [47] Pharmacological work suggests that *S. rebaudiana* can act as a hepatoprotective and cardioprotective agent, mainly because it decreases oxidative stress and helps protect tissues from metabolic damage. [45] Because of these therapeutic roles, along with its low caloric value, stevia is now widely used across nutraceutical, pharmaceutical, and food domains, as a natural sweetener, and more or less as a health-promoting substitutes. [49] More recently, researchers have been focusing on stevia in functional foods, herbal formulations, and medicinal preparations, since it carries diverse bioactive phytochemicals, and it also shows broad pharmacological properties.[44]

2.4.5 Phytochemical and Spectroscopic studies

Some qualitative alongside quantitative phytochemical analysis on *Stevia rebaudiana* have basically confirmed that alkaloids, phenols, tannins, saponins.[45] In many cases UV-Visible spectrophotometry has been used, for measuring and also characterizing phenolic substances and stevia glycosides found within stevia extracts. When FTIR was applied for *S. rebaudiana* extract characterization it showed functional groups like hydroxyl, carbonyl and nitril groups. This evidence validated that various phytochemical constituents are present.[48] Moreover, spectral fingerprinting via UV and FTIR methods has made a big difference for phytochemical profiling for the analytical identification of stevia leaf metabolites and sweet diterpene glycosides.[44] So overall, phytochemical and spectroscopic studies are useful for the medical and pharmacological relevance of those bioactive molecules which are present in *Stevia rebaudiana* [45]

2.4.6 Conclusion

Stevia rebaudiana is widely seen as a medicinally and commercially relevant plant, mainly because it has a relatively rich phytochemical composition and it is used often as a natural low-calorie sweetener, across the pharmaceutical, nutraceutical, and food industries.[43] Steviol glycosides like stevioside and rebaudioside A are the ones that are responsible for most of the therapeutic effects, and the sweetening impact, that people associate with stevia.[46] A lot of pharmacological investigations have pointed out that *S. rebaudiana* shows antidiabetic and antioxidant potential, so, it becomes an interesting option for handling metabolic imbalance and oxidative stress related disorders.[47]

2.5 Introduction to Phytochemicals

Phytochemicals are naturally found bioactive compounds made by plants. In most cases, primary metabolites help with growth and development, while secondary metabolites seem more about ecological adaptation and medicinal use. [50] Secondary metabolites like alkaloids, flavonoids, terpenoids, glycosides and phenolic compounds have a key role in plant defense mechanisms. They help plants fight pathogens, insects, and environmental stress conditions [51] A lot of medicinal plants owe their pharmacological potential, including antioxidant, antimicrobial, anti-inflammatory, and therapeutic effects, to biologically active secondary metabolites that are already present. [50] Because herbal medicine and natural drug discovery keep getting more attention, scientific interest in phytochemicals has also grown. This is mostly due to their therapeutic value and their pharmaceutical relevance [51]

2.5.1 Alkaloids

Alkaloids are nitrogen containing metabolites that occur naturally, and they tend to have strong physiological as well as pharmacological effects, they show up in many medicinal plants [52]. In plants, the production of alkaloids usually starts from amino acids like lysine, tyrosine, tryptophan, and ornithine. This kind of process goes through enzyme mediated metabolic pathways [53]. Medicinal plants seem to make alkaloids for a defensive chemistry, almost like a shield against trouble. They function as protective agents against microbial threat, herbivores, and also environmental pressure [52].

Some therapeutically valuable alkaloid examples are morphine, quinine, atropine, nicotine, and berberine. These compounds are linked with analgesic, antimalarial, antimicrobial and anticancer actions. The antimicrobial activity that we see with alkaloids is often connected to things like blocking nucleic acid synthesis, breaking microbial cell membranes, and interfering with protein formation [54]. Because alkaloids have such a broad biological and pharmacological spectrum, they've become pretty important in today's modern medicine, natural medicine, and drug discovery research too [52].

2.5.2 Phenolic Compounds and Polyphenols

Phenolic compounds are this diverse set of plant secondary metabolites, usually having one or more hydroxyl groups that sit on aromatic benzene rings, and they get grouped broadly into simple phenols as well as polyphenols. [55] Polyphenolic compounds, like flavonoids, phenolic acids and tannins, are often described as showing strong antioxidant effects because they can donate hydrogen atoms, or sometimes electrons, to unsteady free radicals, basically in a sacrificial manner, [56] which helps keep things from spiralling. When you assess antioxidant activity, phenolic compounds are really important for scavenging reactive oxygen species, and they also support the prevention of oxidative damage to cellular biomolecules. [55] At the same time, simple phenols and polyphenols contribute a lot to oxidative stress reduction, mainly by interrupting the free radical chain reactions, and by stabilizing reactive intermediates. [57] Because of that, phenolic antioxidants are often treated as important therapeutic

phytochemicals, since they show protective effects against oxidative stress related conditions and degenerative diseases.[56] And the way phenolic compounds act biologically is mostly linked to their structural traits, especially the count and location of hydroxyl groups, these are the ones that drive how efficient the free radical scavenging becomes.[57]

2.5.3 Tannins

Tannins are polyphenolic compounds, more or less broadly grouped as hydrolysable tannins and condensed tannins based on their chemical structure and how they behave during hydrolysis.[11], [58] A key feature about tannins, is that they can precipitate proteins via strong hydrogen bonding interactions. [60] Also, tannins show antimicrobial effects on different pathogenic microorganisms, mainly because they disturb microbial enzymes, interfere with membrane proteins, and limit substrate availability.[58] Their antioxidant potential is generally connected to the polyphenolic framework, it lets them scavenge free radicals effectively and also lowers oxidative stress.[59] Because of these antioxidant, antimicrobial, and therapeutic roles, tannins are seen as important bioactive parts in medicinal plants and in traditional herbal medicine. [60]

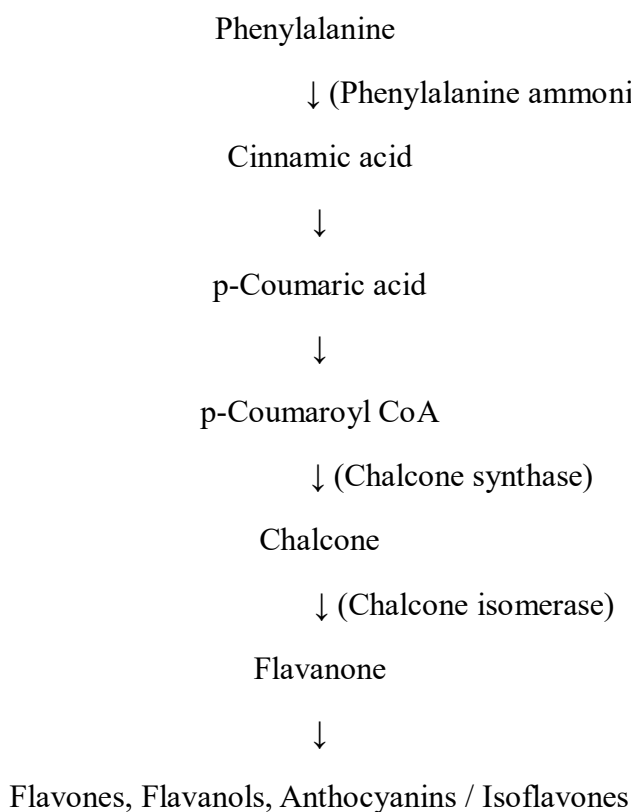
2.5.4. Saponins

Saponins are glycosidic secondary metabolites made up of a hydrophobic aglycone part that is linked to hydrophilic sugar moieties, and that linkage is what gives them their typical surfactant like nature and surface-active behaviour.[61] Because they are amphiphilic in a way, saponins show a strong foaming ability in aqueous solutions and in terms of biological activities, saponins can act as antimicrobial, antifungal, antioxidant, and also immunomodulatory agents. [62] One key reason they show antifungal activity is that they can bind with, or at least interact with membrane sterols in fungal cells, which then leads to higher membrane permeability, and finally to cellular injury. Their anti-inflammatory importance is also tied to blocking inflammatory mediators, and in turn lowering tissue inflammation in different disease related scenarios.

2.5.5 Flavonoids

Flavonoids show strong antioxidant activity, mainly because they donate hydrogen atoms or electrons to neutralize those reactive oxygen species. also they can stop oxidative chain reactions.[65] The antioxidant mechanisms for flavonoids tend to link to metal ion chelation, free radical scavenging, and the hydroxyl groups helping stabilize reactive intermediates, thus minimizing oxidative damage.[63] Flavonoids also have characteristic UV absorption properties, because the conjugated aromatic structures are responsible for this characteristic and they commonly absorb in the UV-A and UV-B regions.[70] A number of flavonoids show important pharmacological effects, for example anti-inflammatory, antimicrobial, anticancer, cardioprotective, and hepatoprotective actions.[64]

2.5.5.1 Biosynthesis of Flavonoid



2.5.6 Terpenoids

Terpenoids are usually broken down into monoterpenes, sesquiterpenes, diterpenes, triterpenes, and tetraterpenes, basically depending on how many isoprene units are present.[67] A lot of terpenoids show up as major parts of essential oils, and they help form the typical aroma, flavour and that of volatile nature found in medicinal as well as aromatic plants.[68] Volatile terpenoid compounds are also important in ecology, they take part in plant protection, and help defend against microbial pathogens or environmental stress conditions.[69] Several terpenoids have antimicrobial activity, they can disturb microbial cell membranes, interfere with enzyme systems, and they also limit microbial growth.[68]. In medicine, terpenoids are important because they bring a range of pharmacological effects, like antioxidant, anticancer, antiviral, antimicrobial, and anti-inflammatory actions.[69]

2.5.6.1 Biosynthesis of Terpenoids

Terpenoid production mostly goes by the mevalonate pathway and the methylerythritol phosphate pathway both of which are present in plants.[71] Isopentenyl pyrophosphate together

with dimethylallyl pyrophosphate work like universal precursor molecules, for making many terpenoid groups.[70] After that, sequential linking of isoprene units (or isoprene pieces) is catalysed by prenyltransferase enzymes and this step gradually builds mono-, sesqui-, di-, and triterpenoid skeletons.[71] Then hydroxylases, cyclases and terpene synthase enzymes convert the terpenoid intermediates even more, so that a wider range of structurally different, biologically active terpenoids can be formed.[67]

2.5.7 Glycosides

Glycosides are bioactive compounds built from a sugar portion, called glycone, which is attached to a non-sugar component known as aglycone by glycosidic bond, and this connection is showing their biological properties as well as their solubility.[69] In general, the pharmacological effects of glycosides depend mostly on what the aglycone part is made of, while the sugar component tends to influence absorption, movement through tissues, and overall bioavailability.[70] Cardiac glycosides are steroidal glycosides, they show a strong action on heart muscle basically by blocking Na^+/K^+ -ATPase activity and they remain therapeutically relevant in some cardiovascular disorders.[69] Steviol glycosides, for instance stevioside and rebaudioside A, are sweet diterpene glycosides obtained from *Stevia rebaudiana* and they are commonly used as natural low calorie sweeteners hence antidiabetic as well as antioxidant, and antihypertensive effects. Glycosides play an important role in medicine since their effects are wide ranging, including cardiogenic, antimicrobial, and anticancer activity as well as other therapeutic uses that show up in pharmaceutical research.[70] Many medicinal plants produce glycosides as important secondary metabolites, and those metabolites help support the pharmacological and therapeutic features.[69] The growing scientific curiosity about glycosides is mostly tied to their broad bioactivity profile, the fact they could be applied in herbal medicine and also in drug development programs.[72]

2.5.8 Sterols and Phytosterols

Plant sterols, usually called phytosterols, are similar in shape to cholesterol, and they mainly include β -sitosterol, stigmasterol and campesterol. Phytosterols have a tetracyclic cyclopentanoperhydrophenanthrene core, but there are small differences in side chain substitution, and also where the double bond is located, these little variations are responsible for structural diversity[73] Sterols are needed to keep plant cell membranes stable, and also they help with permeability and fluidity, basically they control membrane arrangement and cell integrity. Sitosterol and stigmasterol are among the main membrane-associated phytosterols, and they help a lot in stabilizing lipid bilayers and in supporting membrane related physiological roles.[74] Plant sterols show a range of pharmacological activities, such as anti-inflammatory, antioxidant, antimicrobial, anticancer, and cardioprotective effects.[75] Phytosterols are also being considered more often in therapy, mostly because of their cholesterol-lowering action and because they could be used in nutraceutical and pharmaceutical industries.[73] Sterol glycosyltransferases are involved in turning free sterols into sterol glycosides, these glycosides take part in membrane organization, and also help with stress adaptation in plants.[76]

2.5.8.1 Biosynthesis of Sterols

In plants, sterol biosynthesis mostly goes through the mevalonate pathway, where acetyl-CoA gets converted into isoprenoid intermediates, and then later on it leads toward squalene and cycloartenol. Cycloartenol is the main starting point for making different phytosterols and several enzymatic changes then produce structurally varied sterol compounds in plants.[77]The bio synthesis and buildup of phytosterols are crucial for plant growth, the steadiness of membranes, and their ability to adjust to stressful environmental conditions.[74]

2.6 Extraction and Isolation of Phytochemicals

2.6.1 Principles of Phytochemical extractions

In phytochemical extraction, the main idea is really solvent–solute interaction, where bioactive substances into a suitable solvent, based on how they're chemically characterized and on the solubility features. [78] Solvent polarity matters, but it depends upon the extraction efficiency factor, because polar solvents usually draw out hydrophilic phytochemicals, whereas non-polar solvents do much better with lipophilic components. [81] Also, choosing proper solvents is vital to get the highest recovery of both polar and non-polar phytochemicals and, at the same time it supports the stability of the isolated compounds. [79]

2.6.3 Solvents used in phytochemical extraction

Extraction solvents is a crucial step in phytochemical studies, because solvent traits end up shaping both the recovery and the actual makeup of those bioactive compounds pulled out from medicinal plants. [78] In real practice, methanol, ethanol, water-based systems, and hydroalcoholic mixtures are very frequently used for phytochemical extraction because they can dissolve a wide spectrum of secondary metabolites across varied polarities. [80] Solvent polarity matters a lot for the phytochemical yield, and in general polar solvents tend to be better for phenols, flavonoids, and glycosides extraction. [81] Also every extraction solvent has its own set of advantages and little limitations tied to extraction efficiency, toxicity, selectivity, and even how stable the phytochemical constituents remain over time. [79]

2.6.3.1 Methanolic Extraction

Methanolic extraction is often used in medicinal plant research, since methanol seems to work quite well for pulling out phenolics, flavonoids, and other polar phytochemicals.[80] Methanolic extracts, are then used a lot for antioxidant assays, mostly because of their fairly strong ability to scavenge radicals, and also due to the fact that they usually have a rich, or high, set of phenolic constituents.[83]

2.6.3.2 Ethanolic Extraction

Ethanol extraction is seen as a safer solvent because the toxicity levels are lower and it works with both pharmaceutical, and nutraceutical workflows.[79] Ethanol efficiently extract a range of polar bioactive compounds, like phenols, flavonoids, and glycosides from medicinal plants. [78]

2.6.3.3 Water Based Extraction

Water based extraction has been used for quite a while in herbal medicine preparation because water is an accessible and not toxic type of extraction media [83] In practice, aqueous solvents can work really well when you need to pull-out water-soluble phytochemicals like carbohydrates, glycosides and a few phenolic compounds.[80]

2.6.5 Importance of Extraction Methods in Phytochemical Research

In antimicrobial and antioxidant work, choosing the extraction techniques is important, because the extraction efficiency decides how much of the bioactive secondary metabolites are recovered. Those metabolites are related to therapeutic effects.[80] Standardized extraction procedures also matter a lot for UV–Visible and FTIR spectroscopic work, because they improve reproducibility, support consistent phytochemical profiling, and help with validation of herbal extracts.[81] Efficient extraction and concentration method helps a lot with the identification, separation, and characterization of medicinally valuable phytochemicals from plant material. Finally, choosing a best extraction procedure helps a lot with the identification, separation, and characterization of medicinally valuable phytochemicals from plant material.[83]

2.7 Phytochemical Screening Techniques

Qualitative phytochemical screening is a first step analytical approach, it's used to check whether major bioactive secondary metabolites are present in medicinal plant extracts.[85] Usually, standard phytochemical tests are determine for the identification of alkaloids, phenols, tannins, and flavonoids. This is based on typical colour changes, and precipitate formation reaction mixtures.[86] In addition, preliminary screening methods are also used to detect saponins, glycosides, terpenoids, and sterols, these are important phytoconstituents, that are linked to therapeutic activities.[87]

For alkaloids, detection is often done using Dragendorff's, Mayer's, and Wagner's reagents, meanwhile ferric chloride and lead acetate tests are widely used for phenols and tannins.[85] For flavonoids the Shinoda test is a common choice, for saponins the frothing test is applied, and for sterols and terpenoids people often rely on the Liebermann–Burchard test. Together these are major qualitative methods in phytochemical investigations.[86] Overall, preliminary phytochemical analysis matters a lot in medicinal plant research because it helps with the recognition of bioactive compounds, and it also supports further pharmacological, and spectroscopic studies.[87]

2.8 UV-Visible Spectroscopy

2.8.1 Introduction to UV–Vis Spectroscopy

UV–Vis spectroscopy is a technique built around how molecules absorb ultraviolet and visible radiation, which then leads to electronic excitation of chemical substances.[22], [88] This method is commonly used in phytochemical analysis, for identification and quantitative estimation of bioactive compounds that are found inside medicinal plant extracts.[89] When UV light interacts with molecules it excites electrons from lower energy levels to higher energy levels and this produces characteristic absorption spectra.[90]

2.8.2 Principle of UV–Visible Spectroscopy

UV–Visible spectroscopy works on the principle that molecules absorb ultraviolet or visible radiation at certain wavelengths, these match with electronic transitions inside the molecule.[88] For electronic excitation, molecules undergo transitions like $\sigma \rightarrow \sigma^*$, $n \rightarrow \sigma^*$, $\pi \rightarrow \pi^*$, plus $n \rightarrow \pi^*$, and it depends on the bonding type and the functional groups involved.[90] Chromophores are basically the light-absorbing units that are responsible for the characteristic absorption patterns, while auxochromes are the ones that alter absorption intensity and sometimes shift the wavelength a bit.[88] The absorption spectra obtained during UV analysis indicates about the molecular structure and whether phytochemical constituents are present.[89]

2.8.3 Beer–Lambert Law

Beer–Lambert law basically says that absorbance of light by a solution is directly related to the concentration of the absorbing compound, as well as the path length of the light.[89]

$A = \epsilon cl$ here, A means absorbance, ϵ stands for molar absorptivity, c is the concentration, and l represents the path length.[90]

The Beer–Lambert law is the base for quantitative estimating of phytochemicals, and also to figure out the concentration of plant metabolites, present using UV spectroscopy. Quantitative UV spectroscopic analysis is widely used for the estimation of phenols, flavonoids, and other bioactive phytochemicals, in medicinal plant extracts.[91]

2.8.4 Instrumentation of UV–Visible Spectroscopy

In general, the UV–Visible spectrophotometer has a few essential parts which consist of light source, monochromator, sample holder, detector, and the recording system. For light sources, tungsten lamps are usually applied for visible region, and deuterium lamps are used for ultraviolet region, during UV spectroscopic analysis. Then the monochromator selects particular wavelengths, while the cuvette or sample holder holds the sample solution so the absorbance can be measured.[90] Finally the detector transforms the transmitted light into electrical signals, which are then processed and finally shown as absorption spectra by the recording system.[91]

2.9 FTIR Spectroscopy

2.9.1 Introduction and Principle of FTIR Spectroscopy

Fourier Transform Infrared spectroscopy is an analytical method that depends on the absorption of infrared radiation by molecules, and this causes vibrational excitation in chemical bonds found inside a compound. [92] When infrared light interacts with bond systems, it gives rise to a recognizable absorption pattern and those patterns are often useful for understanding the chemical composition of phytoconstituents. [93] FTIR spectroscopy tends to be widely applied in phytochemical investigations, mostly because it supports fast identification and also the characterization of functional group motifs in medicinal plant extract. [94]

2.9.2 Functional Group Identification and Interpretation of FTIR Spectra

FTIR spectroscopy makes it easier to analyze functional groups by examining characteristic absorption peaks, which show up due to stretching and bending movements of molecules. [92] Among the functional groups that are commonly noticed in medicinal plant extracts are O–H, C=O, C–O, and N–H and each one shows up within its own typical absorption zone in the infrared range. [95] For interpreting FTIR spectra, researchers usually match the absorption peaks to phytochemical components like phenols, flavonoids, glycosides, and proteins. [93] Overall, the distinctive FTIR absorption bands give useful insight on structural traits, and also on the chemical character of those bioactive plant metabolites. [94]

2.9.3 Applications of FTIR Spectroscopy in Medicinal Plant Analysis

FTIR spectroscopy is extensively used in the analysis of medicinal plant extracts, mostly for identifying and characterizing bioactive phytochemicals. [94] This method works well for spotting phenols, flavonoids and glycosides, by looking at the absorption peaks that come from specific functional groups. [92] Spectral fingerprinting using FTIR gives a singular chemical profile for plant extracts and helps in phytochemical characterization and also in authentication studies. [93] In addition FTIR analysis is useful in quality control and standardization of herbal drugs, because it makes it easier to check consistency and purity in the medicinal plant preparations. [92]

Chapter 3: - METHODOLOGY

3.1 EXTRACTION OF SUNDRIED LEAVES

3.1.1 Preparation of Plant Extract

Leaves from four chosen medicinal plants were collected from the nursery of Delhi Technological University, Delhi and then used for phytochemical analysis. The collected leaves were washed well with distilled water, so that dust and other impurities could be removed, and after that the plant samples were placed for sun drying for about two days, under natural sunlight, until the moisture was reduced enough. Once they were fully dried, 5 g of the leaves were weighed and grind into a fine powder, using a grinder.

For making the aqueous extract, 1 g of the dried powdered plant material was put into a conical flask with 100 ml of distilled water, and then boiled for about 3 minutes, mainly so the secondary metabolites can come out more easily. After this heating step, the flask was sealed with aluminium foil and kept without touching until the mixture cooled down to room temperature, or something close to that. The extract was then centrifuged at 4000 rpm, at 4°C for 10 minutes. Next, the upper supernatant was collected carefully and kept safely, so it can be used later for both qualitative and quantitative estimation of phytochemicals including alkaloids, phenols, tannins, saponins, flavonoids, quinones, terpenoids and sterols.



Fig 1: - Figure shows steps for the extraction of phytochemicals

3.2 EXTRACTION OF FRESH LEAVES

3.2.1 Preparation of Plant Extract

Fresh leaves from four medicinal plants were collected from the nursery area of Delhi Technological University, Delhi for phytochemical screening. The collected samples were rinsed properly with distilled water, in order to remove the dust and other surface contaminants that were hanging around. Then about 5 g of fresh leaves was weighed, and later crushed in a mortar and pestle until it turned into a thick, semi solid paste, although it was not completely uniform, slightly rough here and there.

For the extract preparation, roughly 1 g of the crushed plant material was transferred into a conical flask containing 100 ml of distilled water. The flask was kept undisturbed for about 3-4 minutes, so that the bioactive constituents could seep out into the solvent. Once a visible colour change showed up in the solution, the extract was centrifuged at 4000 rpm at 4 °C for 10 minutes. After that, the supernatant was separated carefully and kept aside, it would be used later for qualitative as well as quantitative evaluation of secondary metabolites including alkaloids, phenols tannins, saponins, flavonoids, quinones, terpenoids and sterols.

3.3 Qualitative Estimation of Secondary Metabolites

For the qualitative phytochemical screening of the prepared plant extracts, we did the detection of some secondary metabolites like alkaloids saponins, phenols, tannins, flavonoids, terpenoids, sterols and quinones. We used the usual standard biochemical tests. The appearance of a characteristic color shifts, or the formation of a precipitate, generally pointed toward the presence of certain phytoconstituents.

3.3.1 Alkaloids detection (Wagner's Test)

Preparation: Wagner's reagent was made by dissolving 1.27 g iodine and 2 g potassium iodide in 100 ml of distilled water, and stirred until it looked uniform.

Procedure: Take 2 ml of the plant extract, then add only a few drops of Wagner's reagent to it.

Observation: A reddish-brown precipitate appeared; this was taken as evidence of alkaloids.

3.3.2 Saponins detection (Foam test)

Procedure: 2 ml plant extract was shaken vigorously for around 30 seconds, then it was left to stand quietly for a while.

Observation: Stable froth formation indicated the presence of saponins.

3.3.3 Phenols detection (Ferric chloride test)

Preparation: A 5% FeCl_3 solution + 5 g ferric chloride in 100 ml distilled water.

Procedure: Add 1 ml of plant extract, then treat it with a few drops of the 5% FeCl_3 solution.

Observation: Black coloration was considered confirmation for phenols.

3.3.4 Tannins detection

Preparation: A 0.5% FeCl_3 solution was prepared by dissolving 0.5 g ferric chloride in 100 ml distilled water.

Procedure: Mix 2 ml of plant extract with a few drops of the 0.5% FeCl_3 solution.

Observation: Blue-black colour, was taken as the indication that tannins were present.

3.3.5 Flavonoids detection (Alkaline reagent test)

Preparation: A 2% NaOH solution was prepared by dissolving 2 g sodium hydroxide in 100 ml distilled water.

Procedure: 1 ml plant extract was combined with 2 ml of 2% NaOH solution, after that a few drops of diluted H_2SO_4 was added.

Observation: Yellow colour that turned colourless was taken as flavonoids positive.

3.3.6 Test for Terpenoids (Salkowski Test)

Procedure: 2 ml plant extract was mixed with 2 ml chloroform and heated gently.

After evaporation, 2 ml concentrated H_2SO_4 was added and heated for 2 minutes.

Observation: Black coloration indicated the presence of terpenoids.

3.3.7 Test for Sterols

Procedure: 1 ml plant extract was mixed with 10 ml chloroform followed by concentrated H_2SO_4 .

Observation: Formation of reddish-brown ring indicated the presence of sterols.

3.3.8 Test for Quinones

Preparation: Dilute NaOH solution was prepared in distilled water.

Procedure: 1ml plant extract was treated with dilute NaOH solution.

Observation: Blue-green or red coloration confirmed the presence of quinones.

Sr. No.	Preliminary Test	Protocol	Expected Result
1	Alkaloids	2 ml plant extract + few drops of Wagner's reagent	Reddish-brown ppt formation
2	Saponins	2 ml plant extract shaken vigorously in distilled water	Stable froth formation
3	Phenols	1 ml plant extract + few drops of FeCl ₃	Black colour formation
4	Flavonoids	1 ml plant extract + 2 ml of NaOH followed by diluted H ₂ SO ₄	Yellow colour disappears and becomes colourless
5	Terpenoids	2 ml plant extract + 2 ml chloroform + 2 ml conc. Sulphuric acid	Greyish colour formation
6	Sterols	1 ml plant extract + 10 ml chloroform + conc. Sulphuric acid	Reddish-brown ring formation
7	Quinones	1 ml plant extract + dilute sodium hydroxide solution	Blue-green or red colour formation
8	Tannin	2 ml plant extract + few drops of FeCl ₃	Blue-black colour formation

TABLE: - 1 TEST FOR DIFFERENT PHYTOCHEMICALS

3.5 Quantitative Estimation of Total Phenolic Content by UV–Visible Spectrophotometry

The Folin–Ciocalteu colorimetric method was used, and then the UV–Vis spectrophotometer was involved for the measurement.

Principle

The phenolic compounds which are in the plant extract react with the Folin–Ciocalteu reagent when the medium is alkaline. This reaction forms a blue coloured complex, and the stronger the colour, the more phenolics are present in the sample. The absorbance was recorded at 765 nm using a UV–Visible spectrophotometer.

Reagents Required

- . Folin–Ciocalteu reagent (1:10 dilution)
- . Na_2CO_3 solution
- . Plant extract
- . Distilled water

Procedure

1 mg of plant extract was weighed, then dissolved with diluted Folin–Ciocalteu reagent (1:10). About 5 ml of the prepared FC reagent solution was added into a flask that already had the plant extract in it. The reaction mixture was left undisturbed for 3–4 minutes.

After the incubation period, 4 ml of Na_2CO_3 solution was added into the mixture, and mixed properly. Then the flask was kept in dark conditions for 30 minutes to allow the colour to develop.

Observation

The appearance of a bluish coloration suggested that the phenolic complex had formed in the reaction mixture.

Absorbance Measurement

The absorbance of the prepared sample was measured at 765 nm against a suitable blank solution.

Result Interpretation

If the absorbance readings were higher, it meant a greater phenolic content in the plant extract was present.

3.6 Quantitative Estimation of Total Alkaloid Amount by UV–Visible Spectrophotometry

To measure the total alkaloid amount in the plant sample, an ethanolic extraction was done, then the extract was analysed by UV–Visible spectrophotometry

Principle

In the plant, alkaloids (the alkaloidal compounds) were first extracted with 70% ethanol solution. These compounds absorb light at 460 nm and absorbance was taken for the analysis.

Reagents Required

- . 70% Ethanol
- . Dried plant sample
- . Distilled water

Procedure

A small piece of dried plant material, 0.1 g, was weighed and then stirred with about 10 ml of a 70% ethanol solution. It was kept undisturbed, for roughly 30–40 minutes, just to give enough time for the alkaloids to be leached out into the plant solution.

Afterward, the extract was filtered in order to get a clear liquid, and only then the spectrophotometric reading was carried out.

Observation

If a colored ethanolic extract appeared, it implied that alkaloidal constituents were indeed extracted from the plant sample.

Absorbance Measurement












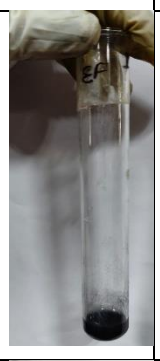




The absorbance of the prepared ethanolic extract was recorded at 460 nm wavelength, with a suitable blank for comparison.

Result Interpretation

In general, absorbance being higher meant the plant extract contained more alkaloids, so greater absorbance values corresponded to a higher alkaloid content.

CHAPTER 4: -OBSERVATION AND RESULTS: -

4.1 QUALITATIVE ANALYSIS OF AQUEOUS SOLUTION OF DRY AND FRESH EXTRACTS: -

PHYTOCHEMICALS	FRESH LEAVES	FRESH FLOWER	SUN DRIED LEAVES	SUN DRIED FLOWER	RESULT
ALKALOID					Red -brown ppt indicate presence of alkaloid
FLAVONOID					Yellow colour does not change to colourless
PHENOL					Black colour indicates presence of phenols
TERPENOID					No greyish colour shows absence of terpenoids







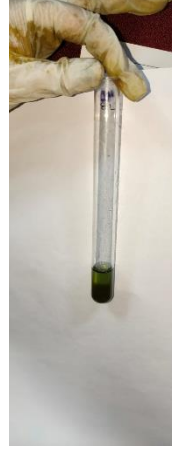

























TANNINS					Blue black colouration indicates tannins are present
STEROLS					Light bluish colouration shows presence of sterols
QUINONES					Reddish colouration shows indication of quinones in fresh but not in dried sample
SAPONIN					Froth formation indicate presence of saponin in fresh flower

TABLE 4.1: - PHYTOCHEMICAL ANALYSIS OF *Euphorbia milli*

PHYTOCHEMICALS	FRESH LEAVES	FRESH FLOWER	SUN DRIED LEAVES	SUN DRIED FLOWER	RESULTS
ALKALOID					Reddish brown ppt indicates presence of alkaloid
FLAVONOID					Yellow colour becomes colourless which shows presence of flavonoid
PHENOL					No black colouration shows absence of phenols
TERPENOID					Greyish colouration shows presence of terpenoid in flower but not in leaves




















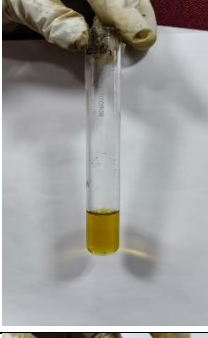

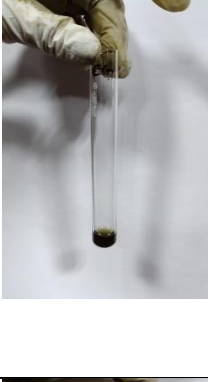


TANNINS					No blue-black colour formation shows indication tannins are absent
STEROLS					Bluish colouration shows presence of phenols
QUINONES					Reddish colour formation shows presence of quinones in fresh flower
SAPONIN					No froth formation indicates absence of saponin

TABLE 3: - PHYTOCHEMICAL ANALYSIS OF *Matricaria chamomilla*

PHYTOCHEMICALS	FRESH LEAVES	SUN DRIED LEAVES	RESULTS
ALKALOID			No reddish-brown precipitate indicates absence of alkaloid
FLAVONOID			Yellow colour does not turn into colourless which indicates absence of flavonoid
PHENOL			Black colouration shows indicates of phenols
TERPENOID			No greyish colour formation indicates absence of terpenoids




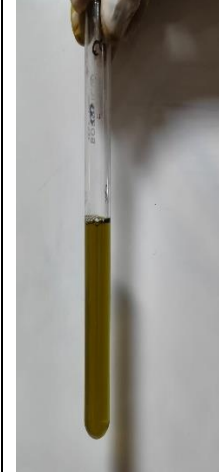

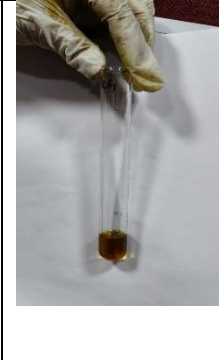










TANNINS			Blue black colouration shows presence of tannins
STEROLS			No bluish colouration which shows absence of sterols
QUINONES			Reddish colouration indicates presence of quinones
SAPONIN			Froth formation indicates presence of saponin

TABLE 4: -PHYTOCHEMICAL ANALYSIS OF *Parthenium hysterophorus*

PHYTOCHEMICALS	FRESH LEAVES	SUN DRIED LEAVES	RESULTS
ALKALOID			Light reddish brown ppt which indicates absence of alkaloid
FLAVONOID			Yellow colour gets colourless which indicates presence of flavonoid
PHENOL			Black colour formation indicates presence of phenols
TERPENOID			Black colour shows indicates presence of terpenoid






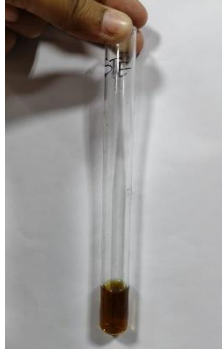


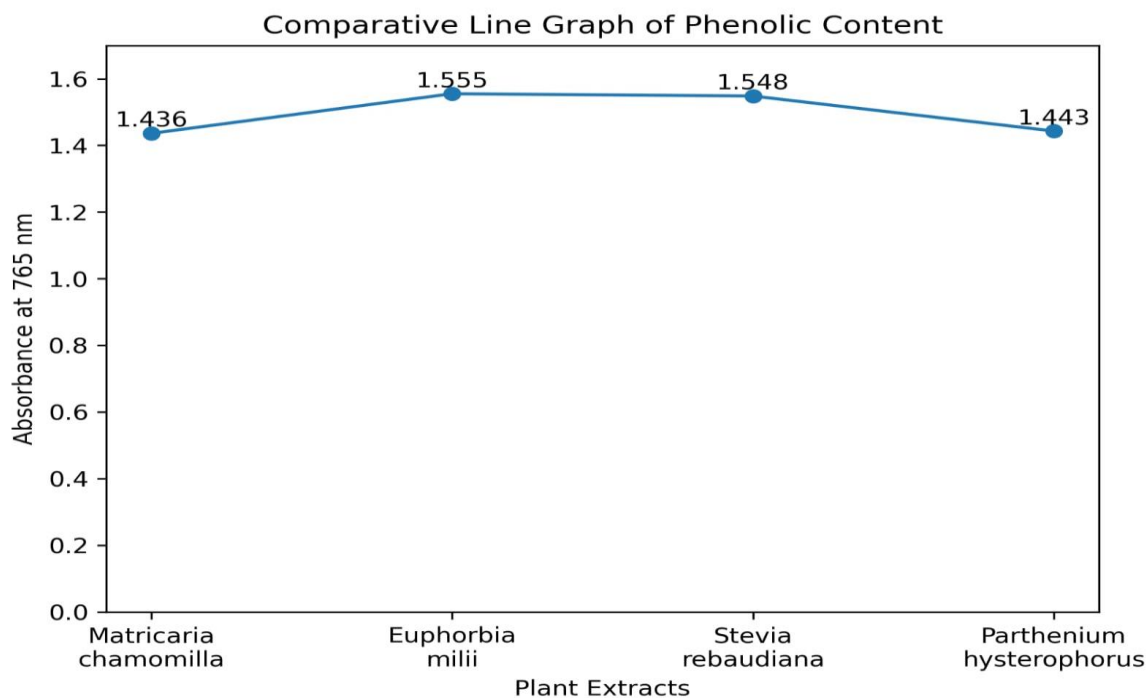
TANNINS			Blue black colouration indicates presence of tannins
STEROLS			Dark bluish colour observed which indicates presence of sterols
QUINONES			Reddish colour formation which indicates presence of quinones in fresh sample
SAPONIN			Froth formation indicates presence of saponin

TABLE: -5 PHYTOCHEMICAL ANALYSIS OF *Stevia rebaudiana*

4.2 QUANTITATIVE ANALYSIS: -

a. UV – VISIBLE SPECTROPHOTOMETER ANALYSIS FOR PHENOLIC CONTENT



Sample Code	Plant Name	Absorbance at 765 nm
CL	<i>Matricaria chamomilla</i>	1.436
EL	<i>Euphorbia milii</i>	1.555
STE	<i>Stevia rebaudiana</i>	1.548
CG	<i>Parthenium hysterophorus</i>	1.443

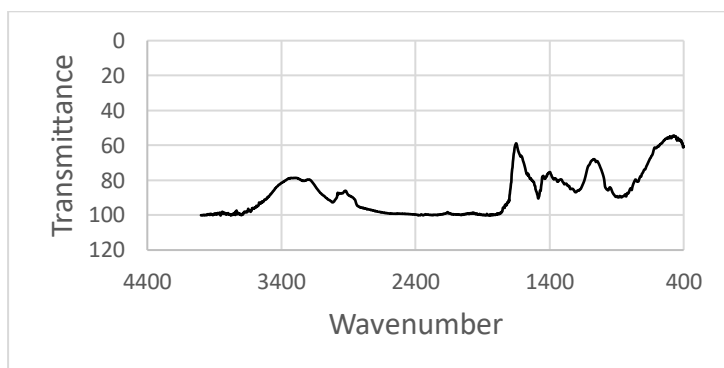
Table 6: - Comparative Absorbance Analysis at 765 nm

Interpretation of Graph

The graph showed an increase in absorbance values corresponding to the phenolic content present in different plant extracts at 765 nm. Among the selected medicinal plants, *Euphorbia milii* exhibited the highest absorbance value (1.555), indicating maximum phenolic content, whereas *Matricaria chamomilla* showed the lowest absorbance value (1.436). The results confirmed the presence of phenolic compounds in all four plant extracts.

b. QUANTITATIVE ANALYSIS OF FTIR

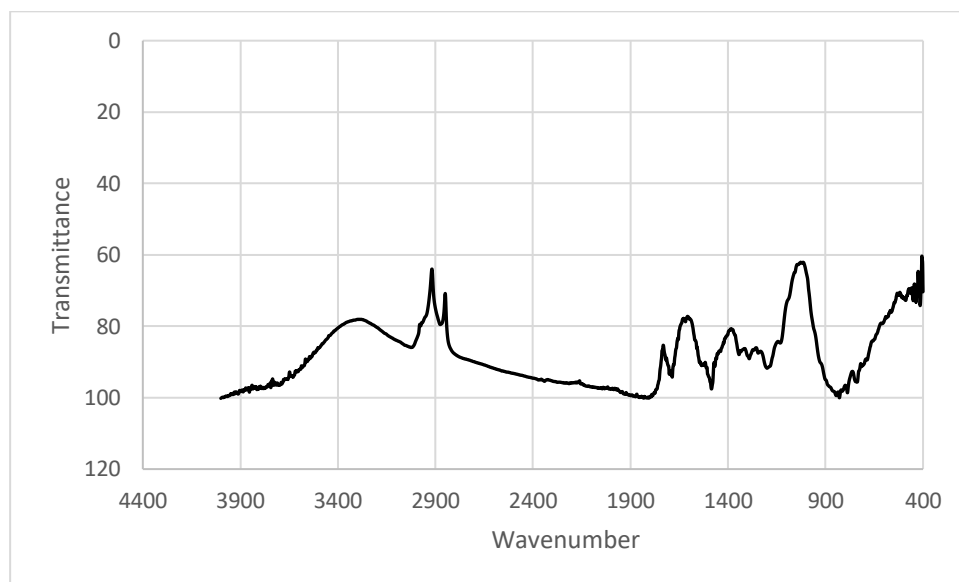
1. Chamomile (*Matricaria chamomilla*)



Peak (cm ⁻¹)	Region	Functional Group	Possible Phytochemical Indication
3400–3300 cm ⁻¹		O–H stretching vibration	Presence of alcohols and phenolic compounds indicating phenols and flavonoids
2950–2850 cm ⁻¹		C–H stretching of alkanes	Indicates aliphatic compounds and terpenoid structures
1700–1600 cm ⁻¹		C=O stretching vibration	Suggests carbonyl compounds such as ketones, aldehydes and flavonoids
1600–1500 cm ⁻¹		C=C aromatic stretching	Indicates aromatic rings commonly present in phenols and flavonoids
1450–1370 cm ⁻¹		C–H bending vibration	Presence of methyl and methylene groups
1250–1050 cm ⁻¹		C–O stretching vibration	Indicates alcohols, ethers and phenolic compounds
1000–800 cm ⁻¹		C–H bending of aromatic compounds	Suggests aromatic phytoconstituents

Table 7: - Interpretation of possible compound in Chamomile by FTIR graph

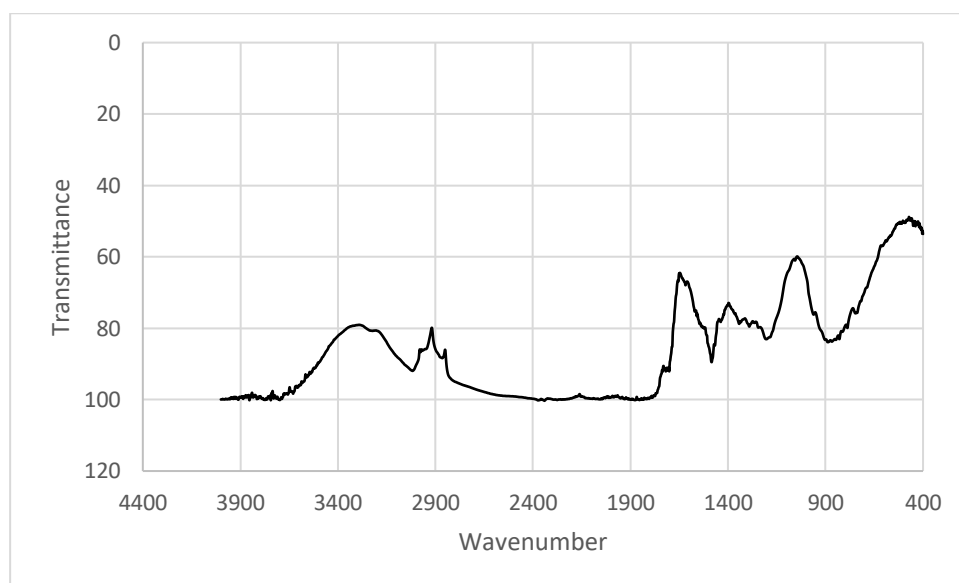
2. *Euphorbia milli*



Peak Region (cm ⁻¹)	Probable Functional Group	Interpretation
3400–3300	O–H stretching	Indicates presence of alcohols or phenolic compounds due to broad hydroxyl absorption peak.
2950–2850	C–H stretching	Represents aliphatic C–H bonds of alkanes, suggesting hydrocarbons or terpenoid constituents.
1650–1550	C=C / N–H bending	May indicate aromatic compounds, amide groups, or alkaloidal constituents.
1450–1380	CH ₂ and CH ₃ bending	Suggests methyl and methylene groups commonly present in plant secondary metabolites.
1250-1050	C–O stretching	Indicates alcohols, ethers, esters, or glycosidic linkages present in phytochemicals.
950-700	Aromatic C–H bending	Suggests aromatic ring structures and phenolic compounds.

Table 8: - Interpretation of possible compounds in *Euphorbia milli* by FTIR

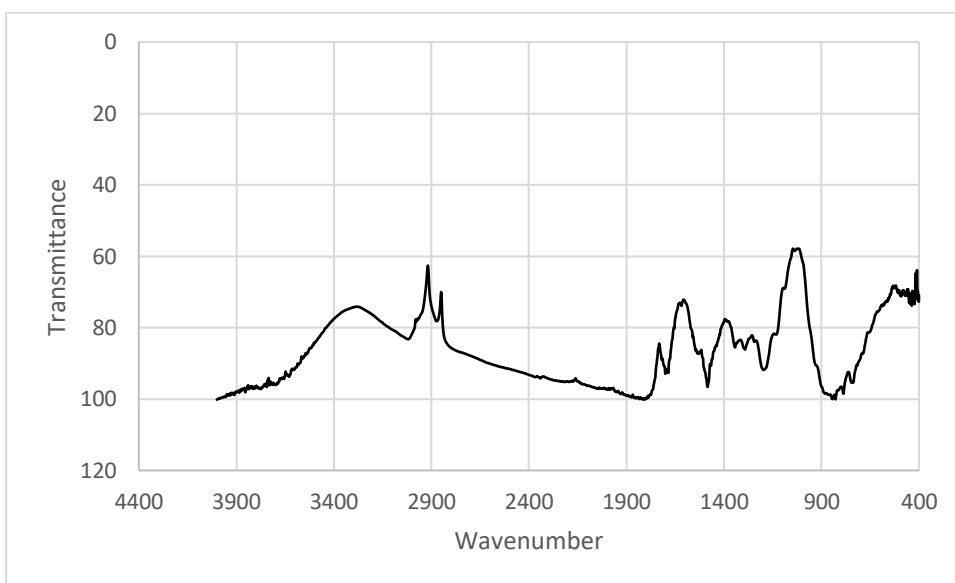
3. *Parthenium hysterophorus*



Peak Region (cm ⁻¹)	Probable Functional Group	Interpretation
3400–3300	O–H stretching	Broad absorption peak indicating presence of alcohols and phenolic compounds.
2950–2850	C–H stretching	Suggests aliphatic hydrocarbons and terpenoid constituents.
1650–1550	C=C / N–H bending	Indicates aromatic compounds, alkaloids, or amide-containing phytochemicals.
1450–1380	CH ₂ and CH ₃ bending	Represents methyl and methylene groups present in organic compounds.
1250-1050	C–O stretching	Suggests alcohols, ethers, esters, and glycosidic compounds.
950-700	Aromatic C–H bending	Indicates aromatic ring structures and phenolic derivatives.

Table 9: - Interpretation of possible compounds in *Parthenium hysterophorus* by FTIR

4. *Stevia rebaudiana*



Peak (cm ⁻¹)	Region	Probable Functional Group	Interpretation
3400–3300		O–H stretching vibration	Broad peak indicating presence of hydroxyl groups associated with phenols and alcohols.
2950–2850		C–H stretching	Indicates aliphatic hydrocarbons and terpenoid constituents.
1650–1550		C=O stretching / C=C stretching	Suggests carbonyl compounds, aromatic rings, or conjugated phytochemicals.
1450–1380		CH ₂ and CH ₃ bending	Represents methyl and methylene groups present in plant metabolites.
1250-1050		C–O stretching vibration	Indicates alcohols, ethers, esters, and glycosidic compounds.
950-700		Aromatic C–H bending	Suggests aromatic and phenolic ring structures.

Table 10: - Interpretation of possible compounds in *Stevia rebaudiana*

CHAPTER 5: DISCUSSION: -

The present study successfully demonstrates quantitative and qualitative phytochemical analysis of 4 important medicinal plants, namely *Euphorbia milii*, *Matricaria chamomilla*, *Parthenium hysterophorus*, and *Stevia rebaudiana* by using aqueous extracts and advanced analytical techniques including UV-Vis and FTIR spectroscopic analysis. The phytochemical screening of the medicinal plants has identified the presence of various important phytochemical constituents including alkaloids, phenols, tannins, flavonoids, saponins, terpenoids, sterols, and quinones.

The quantitative assessment has confirmed the variation in phytochemical constituents among the selected plants, highlighting their unique phytochemical composition and medicinal importance. In addition, UV-visible spectroscopy is considered a very useful tool in the identification and quantification of bioactive compound of medicinal plants. Moreover, FTIR spectroscopic analysis of the samples has indicated the presence of functional groups like hydroxyl and carbonyl groups belonging to phenols, flavonoids, glycosides, and other phytoconstituents.

It has been found that those medicinal plants having high content of phenol and flavonoids showed comparatively higher antioxidant activity. Hence, the study can be regarded scientifically as an important source for the validation of the medicinal importance of the selected plants. It can also be concluded that aqueous extraction is a suitable and economic technique for the phytochemical analysis.

REFERENCES: -

- [1] J. Kanmani Bharathi and M. A. S. Prakash, "Phytochemical screening and quantitative analysis, FTIR and GC-MS analysis of *Costus pictus* D. Don ex Lindl. Leaf extracts – A potential therapeutic herb," *Journal of the Indian Chemical Society*, vol. 101, no. 11, Nov. 2024, doi: 10.1016/j.jics.2024.101362.
- [2] V. Mahawar, S. Yaduwanshi, and G. Jain, "Qualitative and quantitative phytochemical screening of hair growth-promoting plants."
- [3] P. Ch, "December (2022) Res.," "Qualitative and quantitative phytochemical screening of hair growth-promoting plants."
- [4] E. Tekman, T. Asgarlı, H. Yuca, A. Atila, Ö. Çeçen, and S. Karakaya, "Exploring Quantitative Biological Major, Trace, and Ultratrace Elements Composition and Qualitative Primary-Secondary Metabolites in Lamiaceae Medicinal Plants from Turkey," *Biol. Trace Elem. Res.*, vol. 203, no. 2, pp. 1188–1201, Feb. 2025, doi: 10.1007/s12011-024-04219-z.
- [5] Pawar SS and Jadhav MG, "Determination of Extractive value and Phytochemical analysis of *Bacopa monnieri* (L)."
- [6] Y. Srivastav, G. Thakur, and S. Tiwari, "Study of Medicinal Plants: *Ocimum Sanctum* & *Tinospora Cordifolia*, Bioresources to Characterize Bioactive Compounds Using Soxhlet Apparatus," *Asian J. Biotechnol. Bioresour. Technol.*, vol. 10, no. 3, pp. 23–44, Jul. 2024, doi: 10.9734/ajb2t/2024/v10i3209.
- [7] C. Godswill Awuchi, "The Biochemistry, Toxicology, and Uses of the Pharmacologically Active Phytochemicals: Alkaloids, Terpenes, Polyphenols, and Glycosides." [Online]. Available: www.journal.ugm.ac.id/v3/JFPS
- [8] R. M. H. Shoker, "A Review Article: The Importance of the Major groups of Plants Secondary Metabolism Phenols, Alkaloids, and Terpenes," *International Journal For Research in Applied Sciences and Biotechnology*, vol. 7, no. 5, pp. 354–358, Sep. 2020, doi: 10.31033/ijrasb.7.5.47.
- [9] P. Sharma, N. Gautam, A. Arora, H. Ghanghoriya, and A. Anurag, "A Review of Phytochemical Constituents and their Pharmacological Activities.," *Frontiers in Health Informatics*, vol. 14, no. 2, pp. 2645–2658, Sep. 2025, doi: 10.63682/FHI2726.
- [10] W. Sun and M. H. Shahrajabian, "Therapeutic Potential of Phenolic Compounds in Medicinal Plants—Natural Health Products for Human Health," Feb. 01, 2023, *MDPI*. doi: 10.3390/molecules28041845.
- [11] A. K. Farha *et al.*, "Tannins as an alternative to antibiotics," Dec. 01, 2020, *Elsevier Ltd*. doi: 10.1016/j.fbio.2020.100751.

- [12] M. Sánchez-Ramos *et al.*, “Therapeutic Value of Plant Saponins, Challenges and Opportunities: A Review,” *Nat. Prod. Commun.*, vol. 21, no. 2, Feb. 2026, doi: 10.1177/1934578X261418409.
- [13] A. Altemimi, N. Lakhssassi, A. Baharlouei, D. G. Watson, and D. A. Lightfoot, “Phytochemicals: Extraction, isolation, and identification of bioactive compounds from plant extracts,” Dec. 01, 2017, *MDPI AG*. doi: 10.3390/plants6040042.
- [14] S. Sagar and M. Bisht, “A Review on Phytopharmacology of medicinal plant *EUPHORBIA MILII DES MOUL*,” *International Research Journal Of Pharmacy*, vol. 12, no. 6, pp. 67–74, Jun. 2021, doi: 10.7897/2230-8407.1206146.
- [15] J. K. Srivastava, E. Shankar, and S. Gupta, “Chamomile: A herbal medicine of the past with a bright future (review),” Nov. 2010. doi: 10.3892/mmr.2010.377.
- [16] S. Kumar, G. Chashoo, A. K. Saxena, and A. K. Pandey, “*Parthenium hysterophorus*: A probable source of anticancer, antioxidant and anti-HIV agents,” *Biomed Res. Int.*, vol. 2013, 2013, doi: 10.1155/2013/810734.
- [17] M. Ajami *et al.*, “Effects of stevia on glycemic and lipid profile of type 2 diabetic patients: A randomized controlled trial.”
- [18] P. Deshmukh Krishni Vidyapeeth *et al.*, “Phytochemicals: Extraction methods, identification and detection of bioactive compounds from plant extracts,” *J. Pharmacogn. Phytochem.*, vol. 6, no. 1, 2017.
- [19] J. Azmir *et al.*, “Techniques for extraction of bioactive compounds from plant materials: A review,” *J. Food Eng.*, vol. 117, no. 4, pp. 426–436, 2013, doi: 10.1016/j.jfoodeng.2013.01.014.
- [20] G. Rajkumar, P. A. H. R. Panambara, and V. Sanmugarajah, “Comparative Analysis of Qualitative and Quantitative Phytochemical Evaluation of Selected Leaves of Medicinal Plants in Jaffna, Sri Lanka,” *Borneo Journal of Pharmacy*, vol. 52, no. 2, pp. 93–103, 2022, doi: 10.33084/bjop. v5i2.3091.
- [21] D. Casoni, S. C. A. Cobzac, and I. M. Simion, “Feasibility of UV–Vis’s spectroscopy combined with pattern recognition techniques to authenticate the medicinal plant material from different geographical areas,” *J. Anal. Sci. Technol.*, vol. 15, no. 1, Dec. 2024, doi: 10.1186/s40543-024-00428-2.
- [22] P. S. Jirvankar *et al.*, “Phytochemical, UV-Visible and FTIR Assessment along with in vitro Antioxidant Activity of Methanolic Extract of *Tephrosia purpurea* Linn Root,” *J. Pharm. Res. Int.*, pp. 26–33, Jan. 2022, doi: 10.9734/jpri/2022/v34i3b35389.
- [23] S. Agarwal, “Phytochemistry and Pharmacological Potential of *Euphorbia Milii*: Integrating Traditional Therapeutic Applications for Anti-Tumor Activities,” *Biomed. J. Sci. Tech. Res.*, vol. 59, no. 5, Dec. 2024, doi: 10.26717/bjstr.2024.59.009371.

- [24] L. Chen *et al.*, “Traditional Herbal Medicine *Euphorbia milii* Des Moul: Research progress in chemical constituents and pharmacological activities,” Sep. 15, 2026, *Elsevier Ireland Ltd.* doi: 10.1016/j.jep.2026.121772.
- [25] D. Kemboi, X. Peter, M. Langat, and J. Tembu, “A Review of the Ethnomedicinal Uses, Biological Activities, and Triterpenoids of *Euphorbia* Species,” Sep. 01, 2020, *MDPI AG.* doi: 10.3390/molecules25174019.
- [26] “Phyto Chemical Investigation and Isolation of some Bioactive Compounds Present in *Euphorbia milii* Cultivated in Iraq and Evaluation of Antiangiogenic Activity of its Ethyl Acetate Fraction.”
- [27] A. Z. Hassan, H. T. M. Sweelam, K. H. Shaker, M. M. Zohair, W. E. Ashour, and H. I. Abd-Alla, “Phytochemical Analysis and Antimicrobial Activity of *Euphorbia milii*,” *Egypt. J. Chem.*, vol. 66, no. 13, pp. 461–473, Dec. 2023, doi: 10.21608/EJCHEM.2023.192476.7575.
- [28] A. Mahendiran, “A Comprehensive Literature Review on Pharmacological effects of *Euphorbia milii* (Crown of Thorns),” *Advance Pharmaceutical Journal*, vol. 8, no. 4, pp. 94–107, 2023, doi: 10.31024/apj.2023.8.4.3.
- [29] T. A. Chohan *et al.*, “Phytochemical profiling, antioxidant and antiproliferation potential of *Euphorbia milii* var.: Experimental analysis and in-silico validation,” *Saudi J. Biol. Sci.*, vol. 27, no. 11, pp. 3025–3034, Nov. 2020, doi: 10.1016/j.sjbs.2020.08.003.
- [30] R. Kaur, Nishtha, and J. Kumar, “Phytochemical screening, antioxidant activity and metal binding studies on floral extracts of *Euphorbia milii*,” *Mater. Today Proc.*, 2023, doi: 10.1016/j.matpr.2023.03.444.
- [31] K. Ashfaq, M. A. Rehman, M. A. Ghaffari, N. Ali, and N. Sohail, “*Euphorbia milii*: Phenolic contents, Cytotoxic, and Antioxidant Prospect,” *Tropical Journal of Natural Product Research*, vol. 6, no. 8, pp. 1219–1222, Aug. 2022, doi: 10.26538/tjnpr/v6i8.10.
- [32] S. Sagar and M. Bisht, “A REVIEW ON PHYTOPHARMACOLOGY OF MEDICINAL PLANT: *EUPHORBIA MILII* DES MOUL,” *International Research Journal Of Pharmacy*, vol. 12, no. 6, pp. 67–74, Jun. 2021, doi: 10.7897/2230-8407.1206146.
- [33] A. El Mihaoui, J. C. G. Esteves Da Silva, S. Charfi, M. E. C. Castillo, A. Lamarti, and M. B. Arnao, “Chamomile (*Matricaria chamomilla* L.): A Review of Ethnomedicinal Use, Phytochemistry and Pharmacological Uses,” vol. 12, no. 4, Apr. 2022, doi: 10.3390/life12040479.
- [34] Ali Esmail Al-Snafi and Lawahidh Fali Hasham, “Bioactive constituents and pharmacological importance of *Matricaria chamomilla*: A recent review,” *GSC*

- Biological and Pharmaceutical Sciences*, vol. 22, no. 2, pp. 079–098, Feb. 2023, doi: 10.30574/gscbps.2023.22.2.0477.
- [35] V. Gupta, P. Mittal, P. Bansal, S. L. Khokra, and D. Kaushik, “Pharmacological Potential of *Matricaria recutita*-A Review,” 2010. [Online]. Available: www.ijpsdr.com
- [36] N. Shakir *et al.*, “Impact of NaCl stress on phytoconstituents and bioactivity of *Matricaria chamomilla*: a multi-analytical approach,” *Sci. Rep.*, vol. 14, no. 1, Dec. 2024, doi: 10.1038/s41598-024-70672-4.
- [37] S. Patel, “Harmful and beneficial aspects of *Parthenium hysterophorus*: An update,” 2011, *Springer Verlag*. doi: 10.1007/s13205-011-0007-7.
- [38] L. Kaur *et al.*, “Comprehensive review on ethnobotanical uses, phytochemistry, biological potential and toxicology of *Parthenium hysterophorus* L.: A journey from noxious weed to a therapeutic medicinal plant,” Dec. 05, 2021, *Elsevier Ireland Ltd*. doi: 10.1016/j.jep.2021.114525.
- [39] D. Chandra Roy and M. Munan Shaik, “Toxicology, Phytochemistry, Bioactive compounds and Pharmacology of *Parthenium hysterophorus*,” *Journal of Medicinal Plants Studies Year*, vol. 1, no. 1, 2013, [Online]. Available: www.plantsjournal.com
www.plantsjournal.com
- [40] J. Iqbal *et al.*, “Phytochemical Investigation, Antioxidant Properties and In Vivo Evaluation of the Toxic Effects of *Parthenium hysterophorus*,” *Molecules*, vol. 27, no. 13, Jul. 2022, doi: 10.3390/molecules27134189.
- [41] J. Ahmad *et al.*, “Organ-Specific Phytochemical Profiling and Antioxidant Analysis of *Parthenium hysterophorus* L.,” *Biomed Res. Int.*, vol. 2018, 2018, doi: 10.1155/2018/9535232.
- [42] Gul, A. Rauf, I. A. Khan, S. M. Alnasser, S. U. A. Shah, and M. M. Rahman, “Phytochemical Analysis and in Vitro and in Vivo Pharmacological Evaluation of *Parthenium hysterophorus* Linn,” *Evidence-based Complementary and Alternative Medicine*, vol. 2022, 2022, doi: 10.1155/2022/6088585.
- [43] K. Marcinek and Z. Krejpcio, “*Stevia rebaudiana* Bertoni: health promoting properties and therapeutic applications,” Mar. 01, 2016, *Birkhauser Verlag AG*. doi: 10.1007/s00003-015-0968-2.
- [44] S. Khilar, A. P. Singh, M. Biagi, and A. Sharma, “An Insight into attributes of *Stevia rebaudiana* Bertoni: Recent advances in extraction techniques, phytochemistry, food applications and health benefits,” *J. Agric. Food Res.*, vol. 10, Dec. 2022, doi: 10.1016/j.jafr.2022.100458.
- [45] L. Wang *et al.*, “*Stevia rebaudiana* Bertoni as a sweet herbal medicine: traditional uses, potential applications, and future development,” 2025, *Frontiers Media SA*. doi: 10.3389/fphar.2025.1638147.

- [46] A. M. Orellana-Paucar, “Steviol Glycosides from *Stevia rebaudiana*: An Updated Overview of Their Sweetening Activity, Pharmacological Properties, and Safety Aspects,” Feb. 01, 2023, *MDPI*. doi: 10.3390/molecules28031258.
- [47] Z. Peng, S. Zhan, X. Yang, S. Huang, H. Huang, and W. Wang, “The progress on stevia (*Stevia rebaudiana* Bertoni): chemical composition, pharmacokinetics, pharmacological effects, safety, applications, and biosynthesis,” 2026, *Frontiers Media SA*. doi: 10.3389/fnut.2026.1728578.
- [48] S. Patel and A. Navale, “The Natural Sweetener Stevia: An Updated Review on its Phytochemistry, Health Benefits, and Anti-diabetic Study,” *Curr. Diabetes Rev.*, vol. 20, no. 2, May 2024, doi: 10.2174/1573399819666230501210803.
- [49] I. P. Schiatti-Sisó, S. E. Quintana, and L. A. García-Zapateiro, “Stevia (*Stevia rebaudiana*) as a common sugar substitute and its application in food matrices: an updated review,” May 01, 2023, *Springer*. doi: 10.1007/s13197-022-05396-2.
- [50] B. M. Twajj and M. N. Hasan, “Bioactive Secondary Metabolites from Plant Sources: Types, Synthesis, and Their Therapeutic Uses,” *International Journal of Plant Biology*, vol. 13, no. 1, pp. 4–14, Mar. 2022, doi: 10.3390/ijpb13010003.
- [51] R. A. Hussein and A. A. El-Anssary, “Plants Secondary Metabolites: The Key Drivers of the Pharmacological Actions of Medicinal Plants,” in *Herbal Medicine*, IntechOpen, 2019. doi: 10.5772/intechopen.76139.
- [52] Z. Adamski, L. L. Blythe, L. Milella, and S. A. Bufo, “Biological activities of alkaloids: From toxicology to pharmacology,” 2020, *MDPI AG*. doi: 10.3390/toxins12040210.
- [53] S. Schramm, N. Köhler, and W. Rozhon, “Pyrrolizidine alkaloids: Biosynthesis, biological activities and occurrence in crop plants,” Jan. 30, 2019, *MDPI AG*. doi: 10.3390/molecules24030498.
- [54] Y. Yan, X. Li, C. Zhang, L. Lv, B. Gao, and M. Li, “antibiotics Review Research Progress on Antibacterial Activities and Mechanisms of Natural Alkaloids: A Review,” 2021, doi: 10.3390/antibiotics.
- [55] J. Dai and R. J. Mumper, “Plant phenolics: Extraction, analysis and their antioxidant and anticancer properties,” Oct. 2010. doi: 10.3390/molecules15107313.
- [56] A. R. Ndhlala, M. Moyo, and J. Van Staden, “Natural antioxidants: Fascinating or mythical biomolecules?,” *Molecules*, vol. 15, no. 10, pp. 6905–6930, Oct. 2010, doi: 10.3390/molecules15106905.
- [57] J. Das, R. Ramani, and M. O. Suraju, “Polyphenol compounds and PKC signaling,” Oct. 01, 2016, *Elsevier B.V.* doi: 10.1016/j.bbagen.2016.06.022.

- [58] K. T. Chung, T. Y. Wong, C. I. Wei, Y. W. Huang, and Y. Lin, "Tannins and human health: a review," *Crit. Rev. Food Sci. Nutr.*, vol. 38, no. 6, pp. 421–464, 1998, doi: 10.1080/10408699891274273.
- [59] J. Serrano, R. Puupponen-Pimiä, A. Dauer, A. M. Aura, and F. Saura-Calixto, "Tannins: current knowledge of food sources, intake, bioavailability and biological effects," *Mol. Nutr. Food Res.*, vol. 53 Suppl 2, no. SUPPL. 2, pp. S310–S329, 2009, doi: 10.1002/MNFR.200900039.
- [60] L. Zhang, Q. Guan, J. Jiang, and M. S. Khan, "Tannin complexation with metal ions and its implication on human health, environment and industry: An overview," Dec. 31, 2023, *Elsevier B.V.* doi: 10.1016/j.ijbiomac.2023.127485.
- [61] "Saponins in Food, Feedstuffs and Medicinal Plants," *Saponins in Food, Feedstuffs and Medicinal Plants*, 2000, doi: 10.1007/978-94-015-9339-7.
- [62] T. Moses, K. K. Papadopoulou, and A. Osbourn, "Metabolic and functional diversity of saponins, biosynthetic intermediates and semi-synthetic derivatives," *Crit. Rev. Biochem. Mol. Biol.*, vol. 49, no. 6, p. 439, Nov. 2014, doi: 10.3109/10409238.2014.953628.
- [63] S. Chen, X. Wang, Y. Cheng, H. Gao, and X. Chen, "A Review of Classification, Biosynthesis, Biological Activities and Potential Applications of Flavonoids," Jul. 01, 2023, *Multidisciplinary Digital Publishing Institute (MDPI)*. doi: 10.3390/molecules28134982.
- [64] M. C. Dias, D. C. G. A. Pinto, and A. M. S. Silva, "Plant flavonoids: Chemical characteristics and biological activity," Sep. 01, 2021, *MDPI*. doi: 10.3390/molecules26175377.
- [65] N. Shen, T. Wang, Q. Gan, S. Liu, L. Wang, and B. Jin, "Plant flavonoids: Classification, distribution, biosynthesis, and antioxidant activity," Jul. 30, 2022, *Elsevier Ltd.* doi: 10.1016/j.foodchem.2022.132531.
- [66] S. Liga, C. Paul, and F. Péter, "Flavonoids: Overview of Biosynthesis, Biological Activity, and Current Extraction Techniques," Jul. 01, 2023, *Multidisciplinary Digital Publishing Institute (MDPI)*. doi: 10.3390/plants12142732.
- [67] A. Masyita *et al.*, "Terpenes and terpenoids as main bioactive compounds of essential oils, their roles in human health and potential application as natural food preservatives," *Food Chem. X*, vol. 13, Mar. 2022, doi: 10.1016/j.fochx.2022.100217.
- [68] Z. Zhang *et al.*, "Hydroxylases involved in terpenoid biosynthesis: a review," Dec. 01, 2023, *Springer*. doi: 10.1186/s40643-023-00656-1.
- [69] A. F. M. Botelho, F. Pierezan, B. Soto-Blanco, and M. M. Melo, "A review of cardiac glycosides: Structure, toxicokinetics, clinical signs, diagnosis and antineoplastic potential," Feb. 01, 2019, *Elsevier Ltd.* doi: 10.1016/j.toxicon.2018.11.429.

- [70] J. Ren, X. Gao, X. Guo, N. Wang, and X. Wang, "Research Progress in Pharmacological Activities and Applications of Cardiotonic Steroids," Jun. 02, 2022, *Frontiers Media S.A.* doi: 10.3389/fphar.2022.902459.
- [71] J. S. Câmara, R. Perestrelo, R. Ferreira, C. V. Berenguer, J. A. M. Pereira, and P. C. Castilho, "Plant-Derived Terpenoids: A Plethora of Bioactive Compounds with Several Health Functions and Industrial Applications—A Comprehensive Overview," Aug. 01, 2024, *Multidisciplinary Digital Publishing Institute (MDPI)*. doi: 10.3390/molecules29163861.
- [72] J. Ren, X. Gao, X. Guo, N. Wang, and X. Wang, "Research Progress in Pharmacological Activities and Applications of Cardiotonic Steroids," Jun. 02, 2022, *Frontiers Media S.A.* doi: 10.3389/fphar.2022.902459.
- [73] S. Darnet, A. Blary, Q. Chevalier, and H. Schaller, "Phytosterol Profiles, Genomes and Enzymes – An Overview," May 19, 2021, *Frontiers Media S.A.* doi: 10.3389/fpls.2021.665206.
- [74] S. I. Aboobucker and W. P. Suza, "Why do plants convert sitosterol to stigmasterol?" Mar. 22, 2019, *Frontiers Media S.A.* doi: 10.3389/fpls.2019.00354.
- [75] K. Schrick, S. DeBolt, and V. Bulone, "Deciphering the molecular functions of sterols in cellulose biosynthesis," May 03, 2012, *Frontiers Research Foundation*. doi: 10.3389/fpls.2012.00084.
- [76] K. Ramirez-Estrada *et al.*, "Tomato UDP-glucose sterol glycosyltransferases: A family of developmental and stress regulated genes that encode cytosolic and membrane-associated forms of the enzyme," *Front. Plant Sci.*, vol. 8, Jun. 2017, doi: 10.3389/fpls.2017.00984.
- [77] R. B. Jäpelt and J. Jakobsen, "Vitamin D in plants: A review of occurrence, analysis, and biosynthesis," May 13, 2013, *Frontiers Research Foundation*. doi: 10.3389/fpls.2013.00136.
- [78] Q. W. Zhang, L. G. Lin, and W. C. Ye, "Techniques for extraction and isolation of natural products: A comprehensive review," Apr. 17, 2018, *BioMed Central Ltd.* doi: 10.1186/s13020-018-0177-x.
- [79] M. Popova and V. Bankova, "Contemporary methods for the extraction and isolation of natural products," Dec. 01, 2023, *BioMed Central Ltd.* doi: 10.1186/s13065-023-00960-z.
- [80] A. Kumar *et al.*, "Major Phytochemicals: Recent Advances in Health Benefits and Extraction Method," Jan. 01, 2023, *MDPI*. doi: 10.3390/molecules28020887.
- [81] D. Wianowska and M. Gil, "Recent advances in extraction and analysis procedures of natural chlorogenic acids," Feb. 15, 2019, *Springer Netherlands*. doi: 10.1007/s11101-018-9592-y.

- [82] C. L. Okolie, T. O. Akanbi, B. Mason, C. C. Udenigwe, and A. N. A. Aryee, "Influence of conventional and recent extraction technologies on physicochemical properties of bioactive macromolecules from natural sources: A review," Feb. 01, 2019, *Elsevier Ltd.* doi: 10.1016/j.foodres.2018.09.018.
- [83] S. Sun *et al.*, "Impact of extraction techniques on phytochemical composition and bioactivity of natural product mixtures," 2025, *Frontiers Media SA.* doi: 10.3389/fphar.2025.1615338.
- [84] C. R. Travis, J. McMaster, and F. Rivas, "Advances in Natural Product Extraction: Established and Emerging Technologies," Apr. 01, 2026, *Multidisciplinary Digital Publishing Institute (MDPI).* doi: 10.3390/molecules31071136.
- [85] J. R. Shaikh and M. Patil, "Qualitative tests for preliminary phytochemical screening: An overview," *Int. J. Chem. Stud.*, vol. 8, no. 2, pp. 603–608, Mar. 2020, doi: 10.22271/chemi.2020.v8.i2i.8834.
- [86] L. Maheshwaran, L. Nadarajah, S. P. N. N. Senadeera, C. B. Ranaweera, A. K. Chandana, and R. N. Pathirana, "Phytochemical Testing Methodologies and Principles for Preliminary Screening/ Qualitative Testing," *Asian Plant Research Journal*, vol. 12, no. 5, pp. 11–38, Aug. 2024, doi: 10.9734/aprj/2024/v12i5267.
- [87] A. Agarwal, A. Kumar, K. Singh, N. Trivedi, K. K. Jha, and M. A. Agarwal, "A review on extraction and phytochemical screening methods," vol. 2, [Online]. Available: www.rphsonline.com
- [88] "(PDF) UV-Visible Spectroscopy." Accessed: May 25, 2026. [Online]. Available: https://www.researchgate.net/publication/368642160_UV-Visible_Spectroscopy
- [89] "94".
- [90] M. S. H. Akash and K. Rehman, "Ultraviolet-Visible (UV-VIS) Spectroscopy," *Essentials of Pharmaceutical Analysis*, pp. 29–56, 2020, doi: 10.1007/978-981-15-1547-7_3.
- [91] "(PDF) Components, Principle and Applications of UV Vis-Spectrophotometer." Accessed: May 25, https://www.researchgate.net/publication/305402942_Components_Principle_and_Applications_of_UV_Vis-Spectrophotometer
- [92] A. A. Ismail, F. R. van de Voort, and J. Sedman, "Chapter 4 Fourier transform infrared spectroscopy: Principles and applications," *Techniques and Instrumentation in Analytical Chemistry*, vol. 18, no. C, pp. 93–139, Jan. 1997, doi: 10.1016/S0167-9244(97)80013-3.
- [93] B. Van Eerdenbrugh and L. S. Taylor, "Application of mid-IR spectroscopy for the characterization of pharmaceutical systems," Sep. 30, 2011, *Elsevier B.V.* doi: 10.1016/j.ijpharm.2010.12.011.

- [94] H. Tiernan, B. Byrne, and S. G. Kazarian, “ATR-FTIR spectroscopy and spectroscopic imaging for the analysis of biopharmaceuticals,” Nov. 05, 2020, *Elsevier B.V.* doi: 10.1016/j.saa.2020.118636.
- [95] A. Borba and A. Gómez-Zavaglia, “Infrared spectroscopy: an underexploited analytical tool for assessing physicochemical properties of food products and processing,” Feb. 01, 2023, *Elsevier Ltd.* doi: 10.1016/j.cofs.2022.100953.

Certificate -ICBTBI-26



Scopus®

**CERTIFICATE
OF PRESENTATION**



SOCIETY FOR
EDUCATION

**International Conference on Biomedical Technologies and
Bioinformatics (ICBTBI-26)**

17th May 2026 | Hyderabad - India

This is to certify that.....**Akansha**.....affiliated with
.....Delhi Technological University, New Delhi, India.....has presented a paper titled
.....**Role of Bacopa monnieri and Bacosides in modulating Alzheimer disease pathophysiology**.....
.....at the conference organized by
the Society For Education (SFE) held on 17th May 2026 at Hyderabad - India



Dr. James Crusoe
Dr. James Crusoe
President, SFE



Shraddha Srinath
Shraddha Srinath
Convener





DELHI TECHNOLOGICAL UNIVERSITY
(Formerly Delhi College of Engineering)
Shahbad Daultpur, Main Bawana Road, Delhi-42

PLAGARISM VERIFICATION

Title of Thesis: Comparative Qualitative and Spectroscopic Evaluation of Phytochemicals in Medicinal Plants

Total Pages: 58

Name of the Scholar: Chetna Rani & Akansha

Supervisor: Dr. Navneeta Bharadvaja

This is to report that the above thesis was scanned for similarity detection.

Process and outcome are given below:

Software used: Turnitin

Similarity Index: 7%





Total word count: 13222

Date: 25/05/2026




7% Overall Similarity

The combined total of all matches, including overlapping sources, for each database.

Match Groups

-  **64 Not Cited or Quoted 7%**
Matches with neither in-text citation nor quotation marks
-  **0 Missing Quotations 0%**
Matches that are still very similar to source material
-  **0 Missing Citation 0%**
Matches that have quotation marks, but no in-text citation
-  **0 Cited and Quoted 0%**
Matches with in-text citation present, but no quotation marks

Top Sources

- 4%  Internet sources
- 3%  Publications
- 6%  Submitted works (Student Papers)

Integrity Flags

0 Integrity Flags for Review

Our system's algorithms look deeply at a document for any inconsistencies that would set it apart from a normal submission. If we notice something strange, we flag it for you to review.

A Flag is not necessarily an indicator of a problem. However, we'd recommend you focus your attention there for further review.