

**Qualitative analysis of phytochemicals in the Flowers of medicinal
plants: In-silico and In-vitro analysis for Disease treatment**

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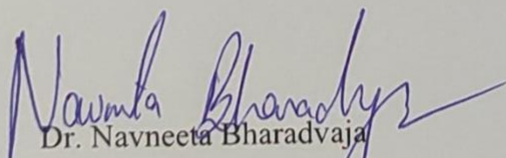
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ABSTRACT

This study looks closely at the phytochemical content of the blooms from five different plant species, including *Chrysanthemum morifolium*, *Dahlia pinnata*, *Gazania rigens*, and *Viola tricolor* var. *hortensis*. The main goal of this study is to determine whether certain phytochemicals, such as tannins, anthocyanins, terpenoids, alkaloids, flavonoids, and terpenoids, are present or absent in these flowers. Through in-silico studies using the SwissTargetPrediction tool to identify the gene targets for the phytochemicals, the thesis also looks at the potential therapeutic efficacy of particular phytochemicals, such as quercetin, apigenin, and isorhamnetin, in the treatment of a number of diseases, including cancer, cardiovascular conditions, pancreatic cancer, Parkinson's disease, and other life-threatening conditions. The results of this study provide vital new information regarding potential medical uses as well as new insights into the phytochemical profiles of the selected plant species. The results also highlight the value of natural ingredients in the development of novel therapeutic agents for the treatment of a range of diseases. The findings indicate a need for further investigation, including clinical trials to evaluate the efficacy and safety of these phytochemicals in human patients, perhaps paving the way for their use in therapeutic settings.

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CHAPTER- 1 INTRODUCTION

1.1 GENERAL INTRODUCTION

The world of plants contains a rich variety of potential therapeutic resources, and the value of using plants for healing is becoming increasingly understood.[1] Plant-based medications have a number of benefits, including being widely available, inexpensive, safe to use, ecologically friendly, and efficient with little adverse effects. Numerous problems have been treated by certain plants by humans for a very long time. Modern scientists are now studying these plants to find new medicines that are effective against diseases like cancer, infections, and liver issues. The World Health Organisation (WHO) recognises that medicinal plants are a valuable source of a wide range of medications. In developed countries, almost 80% of the populace uses medications made from medicinal plants.[2] As a result, we appreciate how valuable plants are. Natural treatments made from plants continue to be in high demand worldwide. Bioactive substances found in medicinal plants have been shown to be useful in treating a variety of human ailments and speeding up the healing process. A medicinal plant is any plant that has components or precursors that are useful for therapeutic purposes.[3] Secondary metabolites as well as essential oils are widely distributed in these plants. While treating existing illnesses rather than preventing them is frequently the main goal when using medicinal plants, it is crucial to remember that phytochemicals—naturally present chemicals in plants—offer a variety of defensive characteristics despite not being necessary for sustenance. Plants contain various amounts of phytochemicals, which can be classified as secondary or primary metabolites. They include tannins, flavonoids, saponins, steroids and alkaloids and they help to give plants their flavour, colour, and scent. They can also protect people from a wide range of ailments, including as arthritis, diabetes, heart disease, cancer, and ageing. The anticancer, antiviral, antifungal, antidiabetic, and antibacterial characteristics of phytochemicals such flavonoids, alkaloids, tannins, saponins, phenols, and others help to prevent a variety of diseases. According to their historical and customary use in various medical systems and their potential to lead to the discovery of novel medications, medicinal plants are carefully chosen for this study. Wide-ranging phytochemical variety is ensured by the integration of distinct plant families and geographical regions. The five chosen therapeutic plants—*Gazania rigens*, *Dahlia pinnata*, *Alcea rosea* (Hollyhock), and

Viola tricolour var. *hortensis*—were subjected to phytochemical screening assays. Numerous types of phytochemicals, such as tannins, saponins, terpenoids, phenols and alkaloids were discovered in the obtained extracts from these plants. By qualitative analysis and the determination of target molecules, the main goal of this study is to investigate the phytochemical makeup and effectiveness of these plants for use as treatment agents for various diseases. Through qualitative analysis, the aim of the following study is identification of bioactive compounds present in these plants and evaluate their potential therapeutic applications. The study seeks to deepen our understanding of these medicinal plants, establish a scientific foundation for their medicinal uses, and contribute to the fields of drug discovery, the validation of traditional medicine, and the potential industrial applications of their bioactive compounds. Ultimately, the main goal of this study is to contribute to the development of effective and alternative treatments of ailments by harnessing the natural compounds derived from medicinal plants.

1.2 PLANT METABOLITES

As a by-product of metabolism, metabolites are molecules that include both intermediate components and end compounds. Metabolites are typically tiny molecules that perform a variety of tasks for an organism. They provide energy, create structural elements, serve as signalling molecules, affect the activity of enzymes, have catalytic capabilities, serve as a defensive mechanism, and interact with other species. Plants create a wide range of organic chemicals, the bulk of which have no direct impact on growth and development.[4] Secondary and Primary metabolites are the two main categories under which plant metabolites fall. For core metabolic functions including respiration and photosynthesis, primary metabolites are essential. They are created through the developmental phase and are essential in sustaining the physiological processes of the organism. Due to their role as intermediates in anabolic processes, which act as the raw materials for crucial macromolecules, these metabolites are regarded as essential to metabolism. vitamins, organic acids and amino acids are a few examples of primary metabolites which are created in huge quantities.[5] Fats and carbohydrates, which are categorised as main metabolites, provide energy for physical activity. Plant secondary metabolites, on the other hand, have unique uses in medicines, food additives,

flavours, and diverse industrial components. Plant cell cultures have been used to study the formation of these secondary metabolites, and organised cultures, in particular root cultures, have shown substantial promise. Organic substances known as secondary metabolites don't directly support an organism's healthy development, growth, or reproduction. While they don't directly affect development and are normally synthesised after the growth period, they might help the organism survive. Particular taxonomic groups of bacteria frequently produce secondary metabolites, which have distinctive chemical structures. Within a chemical family, they are typically encountered as combinations of closely related molecules. The stimulating substances nicotine and caffeine are examples of secondary metabolites. Alkaloids, a different class of secondary metabolites, have therapeutic benefits and are used as analgesics or treatments for a range of illnesses. Secondary metabolites are primarily produced throughout the idiophase, or stationary phase of growth. Steroids, phenolics, alkaloid compounds, dyes, antibiotics, and essential oils are some other instances of secondary metabolites.

1.3 OBJECTIVES

The objectives of this project are

1. To determine the presence of secondary metabolites in the following medicinal plants.
 - *Viola tricolor var. hortensis*
 - *Gazania rigens*
 - *Alcea rosea (Hollyhock)*
 - *Dahlia pinnata*
 - *Chrysanthemum morifolium*
2. To target various disease by these phytochemicals using in silico tools.

1.4 ORGANIZATION OF THESIS

Chapter 1 represents general introduction of medicinal plant and Plant metabolites.

Chapter 2 is a review of literature which contains a broad knowledge about the overview of secondary metabolites Flavonoids, tannins, terpenoids, Anthocyanins, Anthraquinones and Alkaloids. It also covers the information about five medicinal plants *Viola tricolor var. hortensis*, *Gazania rigens*, *Alcea rosea*, *Dahlia pinnata*, and *Chrysanthemum morifolium*.

Chapter 3 is a proposed methodology for the project and briefs about the protocols for preliminary tests for secondary metabolites and how can predictions be made for the use of various phytochemicals for treatment of diseases

Chapter 4 contains result, discussion and conclusion.

CHAPTER 2. REVIEW OF LITERATURE

2.1 MEDICINAL PLANTS

Humans have benefited greatly from both curative and preventive medical treatments thanks to medicinal plants. Many vital bioactive chemicals have been extracted from these plants. Surprisingly, traditional medicine and its products have increasingly replaced all other forms of healthcare for almost 80% of the population of the world, especially in developing nations. To address their health concerns, people in many poor nations frequently combine traditional medicine with modern medical procedures.[6] Traditional medicines serve as the main natural cures available to isolated rural people in poor countries and are frequently more affordable than contemporary medications. The world's most traditional kind of medicine, medicinal plants have been employed for thousands of years in many different traditional medical systems.[7]

The important benefit of medicinal plants has long been acknowledged by traditional medical practises in various civilizations across the globe. These plants include a wide range of bioactive substances, including as terpenoids, flavonoids, and alkaloids. These substances have a variety of pharmacological qualities, including anti-inflammatory, antibacterial, antioxidant, and anticancer effects. In order to conduct a qualitative analysis for this study, we have selected five unique medicinal plants (*Viola tricolor* var. *hortensis*, *Gazania rigens*, *Alcea rosea*, *Dahlia pinnata*, and *Chrysanthemum morifolium*).

Table 1. Ethnobotanical details of chosen medicinal plant species for phytochemical investigation and their therapeutic advantages.

S.No.	PLANT SPECIES	COMMON NAME	PART USED	PRESENT PHYTOCHEMICALS	MEDICINAL BENEFITS
1	<i>Alcea Rosea</i>	Hollyhock	Flowers	alpha-phellandrene, Cholesterol, β -sitosterol, stigmasterol,	To cure wounds, burns and cuts. Used as an herbal tea or decoction

					to treat internal inflammation
2	<i>Viola tricolor</i> <i>var. hortensis</i>	Pansy	Flowers	Quercetin, Glycosides, Alkaloids	Treat minor seborrheic skin conditions include acne, cradle cap, dandruff, and itching. Pansy is used to treat internal skin conditions.
3	<i>Dahlia pinnata</i>	Dahlia	Flowers	flavonoids, tannins saponins and phenols	Treat skin fissures, rashes, and grazes that are infected. prevents constipation
4	<i>Gazania rigens</i>	Gazania	Flowers	Aliphatic triterpenoids, fatty acids, flavonoids,	preserve cardiovascular health, boost immunity, and improve fitness and strength
5	<i>Chrysanthemum morifolium</i>	Guldavari	Flowers	zeaxanthin, 13-cis- β - carotene, β - cryptoxanthin, 9-cis- β -carotene.	Anti-inflammatory, antipyretic, and sedative

2.1.1 Botanical name: *Alcea Rosea*



Fig 1. Alcea Rosea

Classification-

Kingdom: Plantae

Phylum: Tracheophyta

Class: Magnoliopsida

Order: Malvales

Family: Malvaceae

Genus: *Alcea*

Species: *Alcea Rosea*

The gorgeous Malvaceae family member *Alcea Rosea*, sometimes known as the hollyhock, is a biennial or perennial flowering plant. This tall, imposing plant, which is native to both Asia and Europe, is prized for its colourful, brilliant blossoms. Large, saucer-shaped blossoms of the *Alcea Rosea* occur in a variety of hues, including pink, purple, white, yellow, and red. In gardens, flowers are grouped in towering spikes along the hardy stalks, producing an impressive aesthetic show. It has several uses and medical qualities in addition to its visual appeal. The plant has been used for its demulcent and emollient effects in traditional herbal medicine. It can be used to treat coughing and sore throat symptoms and has calming effects on the respiratory system. *Alcea Rosea* roots are useful in natural medicine since they also contain substances that may have antibacterial activity. With its petals utilised as an edible ornament or flavouring in some dishes, this amazing flower species has also made its way into

culinary traditions. The Alcea Rosea continues to enthrall gardeners, herbalists, and nature lovers with its beauty, variety of uses, and potential health benefits.

Medicinal uses of Hollyhock:

1. **Effects against inflammation:** The plant has anti-inflammatory chemicals that may be helpful in treating illnesses including arthritis and joint pain.
2. **Support for the immune system:** Antioxidants found in hollyhocks can help boost immunity and guard against oxidative stress.
3. **Mucous membrane-soothing abilities:** Hollyhock possesses mucous membrane-soothing abilities, making it beneficial for conditions like dry coughs, sore throats, and respiratory allergies.

2.1.2 Botanical name: *Viola tricolor var. hortensis*



Fig 2. Viola tricolor var. hortensis

Classification-

Kingdom: Plantae

Phylum: Tracheophyta

Class: Magnoliopsida

Order: Violales

Family: *Violaceae*

Genus: *Viola*

Species: *Viola tricolor var. hortensis*

The usual height of *Viola tricolor*, which has a ramping and creeping growth tendency, is little more than 15 cm (6 inches). Its blooms, which have a diameter of roughly 0.59 inches or 1.5 centimetres (0.59 cm), bloom in short grassland sections of farms and wastelands, mostly on acidic or neutral soils. The plant loves some shade. Rhizome-like rootlets make up the tiny rootlets that make up its root system. The stem of *Viola tricolor*, often referred to as the "acoli stem," is the base from which the leaves and flowering stalks emerge and stays near to the ground. The stem branches out and is normally smooth, but it occasionally has a downy texture. *Viola tricolor* does not develop a leaf rosette at the base like some other violet species like *Viola hirta*. Instead, the stem of this plant has its leaves placed alternately. The stalked leaves have serrated edges and an oval, oblong, or lanceolate form. The stipules, especially those on the upper leaves, are frequently well-developed and may have a structure resembling a palm. *Viola tricolor* has single flowers that are high and positioned laterally on lengthy peduncles. They come out of aerial stems with different-length internodes. The sepals range in length from 10 to 25 mm and are never longer than the corolla. Purple, blue, yellow, and white are just a few of the colours that the corolla itself can exhibit. It frequently has a two-tone appearance that combines purple and yellow tones. Particularly popular is the tricolour variety, which combines shades of yellow, white, and purple. In the Northern Hemisphere, the flowering season for *Viola tricolor* lasts from April through September. The hermaphrodite, self-fertile plants are frequently pollinated by bees.

The development traits of *Viola tricolor* L. vary depending on the subspecies. It might have a short rhizome or none at all, and it can be annual, biennial, or perennial. *Viola tricolor* has up to 30 cm tall stems with oval-shaped, toothed-edged leaves. These alternately placed leaves have rounded bases. Also unique are the plant's strongly lobed, pinnate stipules, which resemble leaf architecture. The *Viola tricolor*'s (1-2.5 cm in diameter) summertime blooms come in a variety of hues, including white, yellow, and various violet tones. The length difference between the petals and sepals is a distinguishing feature of this plant. It has a light aroma, but it's not extremely potent.[8]

Some special traits about the pansy flower are-

1. They are known to have antimicrobial activity
2. The flowers show a great antioxidant nature. [9]

3. Pansies are edible flowers and are hence used in garnishing as well as cake decorations

2.1.3 Botanical name: *Dahlia pinnata*



Fig 3. Dahlia pinnata

Classification-

Kingdom: Plantae

Phylum: Tracheophyta

Class: Magnoliopsida

Order: Asterales

Family: Asteraceae

Genus: *Dahlia*

Species: *Dahlia pinnata*

The magnificent flowering plant *Dahlia pinnata*, also referred to as *Dahlia*, is a constituent part of the Asteraceae family. This herbaceous perennial, which is native to Mexico and Central America, has become well-known all over the world for its alluring blossoms and varied forms. A broad variety of flower sizes and shapes, including single, semi-double, and double variations as well as intricate shapes like cactus, pompon, and waterlily, are displayed by *dahlia pinnata*. The blossoms display a wide range of hues, including, among others, red, yellow, orange, pink, and white. *Dahlia pinnata* gives gardens and floral arrangements a vivid touch with its rich foliage and eye-catching blossoms. This flower species has notable uses and medicinal qualities in addition to its decorative value. Different plant parts have previously been utilised in

traditional medications for the treatment of ailments like inflammation, discomfort, and skin problems. The antibacterial activity of *Dahlia pinnata* has also attracted attention for potential medicinal uses. *Dahlia pinnata* continues to be a popular option for gardeners, flower lovers, and those seeking the natural beauty and potential healing capabilities of this wonderful species thanks to its astonishing diversity and potential health benefits.

Medicinal uses of *Dahlia pinnata*:

1. Antioxidant property:

Flavonoids and polyphenols, two substances found in dahlia, have antioxidant properties. *Dahlia pinnata* contains antioxidants that work to combat damaging free radicals in the body, thereby lowering oxidative stress and averting potential cellular damage.

2. Antimicrobial activity:

Antimicrobial action against a variety of diseases, including bacteria and fungus, has been demonstrated by extracts from *Dahlia*. It may be helpful in herbal treatments for microbial illnesses thanks to these antibacterial characteristics.

3. Anti-inflammatory activity:

The anti-inflammatory qualities of *dahlia pinnata* can aid in reducing bodily inflammation. These characteristics might be useful for treating inflammatory skin disorders, joint discomfort, and arthritis.

2.1.4 Botanical name: *Gazania rigens*



Fig 4. Gazania rigens

Classification-

Kingdom: Plantae

Phylum: Tracheophyta

Class: Magnoliopsida

Order: Lamiales

Family: *Lamiaceae*

Genus: *Plectranthus*

Species: *Gazania rigens*

A well-known perennial herbaceous plant native to Southern Africa is called gazania (*Gazania rigens* L.). Due to its high ornamental value, it is widely cultivated around the world as a garden plant as well as an annual potted flower. A spreading, low-growing, semi-hardy perennial, *Gazania rigens* grows to a height and breadth of roughly 50 cm (20 inches). Throughout the summer, it produces eye-catching yellow composite flowerheads that resemble daisies and displays blue-grey leaves. It acts as an annual in gardens in cooler climates, despite being a perennial herbaceous plant in South Africa and the Mediterranean. Its normal development is restricted to 30 cm (12 inches), creating tufts that are widespread. The majority of the plant's leaves are basal, numerous, narrow, and somewhat lanceolate in shape. Normally whole, they might infrequently have pinnate lobes. The undersides of the leaves are a greyish-white colour, while the upper surfaces are a bright green colour.[10]

Like other composite plants, Gazania also develops flower heads, which are sometimes mistaken for individual blooms. Capitula, or single flower heads, are found at the tips of peduncles just above the leaves. Each capitulum has a central disc made of tubular flowers that is encircled by ligulate blooms that can be a wide range of colours. The most typical flowers are orange-yellow; however, they frequently include black patches at the base of the ligules. The *Arctotis* genus is similar to the herbaceous perennial prize flower, which is a member of the Asteraceae family. This South African native prefers full sun exposure and does best in sandy to moderately well-drained soil. It is an efficient soil protector even if it needs constant hydration. The delicate perennial decumbent branches of the treasure bloom spread out horizontally along the ground. Its leaves, which are silvery green, add to the entire appeal. The flower heads are often bright orange with a black base and resemble daisies. The majority of the year is spent in flowering, and the plant produces egg-shaped seeds with long hairs.

Medicinal uses of Gazania

1. The Gazania extract has shown Antioxidant activity
2. It has an hepatoprotective effect upon CCL4 induced hepatotoxicity
3. It shows a neuroprotective effect from CCL4 induced neurotoxicity

Use of Gazania extracts showed reduction in the serum levels of CCL4 which have been shown to be responsible for hepato-renal toxicity.[11]

2.1.5 Botanical name: *Chrysanthemum morifolium*



Fig 5. Chrysanthemum morifolium

Classification-**Kingdom:** Plantae**Phylum:** Tracheophyta**Class:** Magnoliopsida**Order:** Asterales**Family:** Asteraceae**Genus:** *Chrysanthemum***Species:** *Chrysanthemum morifolium*

This Plant is most abundantly found in the east Asian regions of China and Japan. This plant is known to have a long and noted cultural heritage and is recognized for flowers which are stunning. *Chrysanthemum morifolium*, it has a variety of flower arrangements ranging from semi-double, fully-double and varieties of solitary form. There are a variety of colours available in this species like yellow, white and purple. It has an extensive medicinal use and traditional Chinese medicine uses these flowers for its cooling properties as well as anti-inflammatory and anti-oxidant activity.

Medicinal uses of Chrysanthemum:

1. *Chrysanthemum* has been used for a long time in the liver wellness and offer protection against various liver ailments.
2. *Chrysanthemum* is effective in treating digestive ailments.

2.2 SECONDARY METABOLITES OF PLANTS

Substances often called as secondary metabolites are formulated by plants, they can possess a variety of uses in their growth and interactions with the environment. Secondary metabolites are not directly engaged in the development, or reproduction, in contrast to primary metabolites, which are necessary for fundamental plant processes. Instead, they aid in communication, adaptability, and plant defence.

These tiny molecules affect plants and other living things in a variety of ways. They have the ability to control leaf abscission, fruit development, and blooming. In response to environmental cues, secondary metabolites can also promote perennial growth or

indicate deciduous behaviour. They serve as antimicrobial compounds that shield plants from diseases and have the ability to attract or repel pollinators, herbivores, and other animals. Over 50,000 different secondary metabolites have been discovered so far, most of which are found in the plant kingdom. Alkaloids, phenolics, terpenoids, flavonoids, and many more chemically diverse molecules are among these substances. These secondary plant metabolites' therapeutic properties are used in various modern medications as well as medicinal plants. They may contain anti-inflammatory, antioxidant, antibacterial, and anticancer properties that are advantageous to human health.

Understanding the functions and potential uses of secondary metabolites in plants has important ramifications for a number of disciplines, including pharmacy, agriculture, and ecology. These substances' complex activities are still being uncovered through research, and they may one day be used to create novel medicines, boost crop protection, and better comprehend how plants interact with their environment.[12]

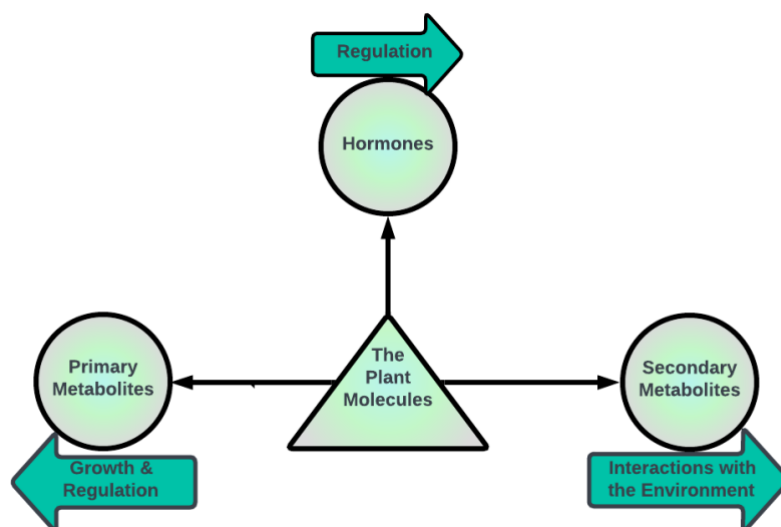


Fig 6. The Major chemicals produced by plants (Hormones, Primary Metabolites & Secondary metabolites)

Different types of secondary metabolites that are made by the plants are: -

2.2.1 FLAVONOIDS

Plants include a group of substances called flavonoids, which can be found in a variety of shapes, including aglycones (flavonoids without connected sugars), glycosides (flavonoids with attached sugars), and methylated derivatives. Flavonoids have two six-membered rings with a total of fifteen carbon atoms, joined by a three-carbon unit that may or may not be a part of a third ring.[13] Common names for these rings are A, B, and C rings. For the A, C, and B rings, the carbon atoms within the rings are identified using standard numbers, and the carbon atoms within the B ring are identified using "primed" numerals. The sites of numerous functional groups and changes inside the flavonoid structure are described using this numbering system. Flavonoids have a wide range of biological functions and are significant in plant physiology, human health, and other applications due to their complex structural makeup. Like other phenolic chemicals, flavonoids have strong anti-oxidant and metal-chelating qualities. They can scavenge free radicals and shield cells from oxidative stress because to these properties. In addition to their ability to act as antioxidants, flavonoids are well known for having a number of positive benefits on human health. They have anti-inflammatory qualities that can help treat associated diseases and lessen inflammation. Additionally, flavonoids have anti-allergic properties that help in the treatment of allergies and allergic responses. They have also been discovered to have hepatoprotective qualities, promoting liver function and guarding against liver injury. Flavonoids have demonstrated antithrombotic action, which implies they can aid in preventing blood clots from forming. They also have antiviral characteristics, which prevent some viruses from reproducing and being active. Last but not least, flavonoids have been studied for their possible anticarcinogenic effects since they may help stop the proliferation of cancer cells. The tremendous potential of flavonoids in enhancing human health and avoiding numerous diseases is highlighted by their varied activities.[13]

2.2.2 TERPENOIDS

Terpenoids, commonly referred to as isoprenoids, are a broad class of organic substances present in all living things. They are distinguished by their construction from isoprene units, and they can be altered in a variety of ways to provide a huge variety of structures and functions. The biggest class of natural products, terpenoids are produced by a variety of organisms, including bacteria, fungi, mammals, and plants.

Oriental herbal treatments frequently incorporate plant terpenoids because of their well-known fragrant qualities. They have also attracted interest for their antibacterial properties, and their ability to fight different infections is currently being extensively researched. Terpenoids are responsible for the distinctive fragrances and flavours of many plants, including the aromas of cinnamon, clove, eucalyptus, and ginger. THC and CBD, two cannabinoids present in Cannabis species, are also categorised as terpenoids. Additionally, several terpenoids have proven to have strong inhibitory effects on the development of the antibiotic-resistant bacteria known as vancomycin-resistant enterococci (VRE). This demonstrates how effective terpenoids could be against microbes.

Terpenoids are significant and fascinating substances in the world of natural products and medicine because they exhibit extraordinary structural diversity and support a variety of biological functions.[14]

2.2.3 ALKALOIDS

Alkaloids are a large group of chemically varied substances that are formed from amino acids and have a heterocyclic ring with a nitrogen atom in it. The main focus of their research is on the distribution, chemical characteristics, and biosynthetic precursors of alkaloids. A lot of research has been done on alkaloids because of their advantageous biological characteristics. Alkaloids, like D-tubocurarine, for instance, have the ability to relax muscles at neuromuscular junctions by inhibiting acetylcholine receptor sites. Alkaloids have been found to have antioxidant properties.[15] To lessen oxidative stress, they can serve as free radical scavengers, bind metal ions, and donate electrons or hydrogen atoms. Leukaemia and Hodgkin's disease are both effectively treated with the use of the alkaloids vinblastine and vincristine, which are produced from the plant *Catharanthus roseus* (Apocynaceae). By destroying or depolymerizing protein microtubules, which are crucial for the formation of the mitotic spindle during cell division, these alkaloids have a chemo preventive impact. Alkaloids' varied features and prospective uses in a number of disciplines, including medicine, pharmacology, and plant biology, are still being uncovered by research.

2.2.4 ANTHOCYANINS

Anthocyanins are a particular class of flavonoids that give different plant parts including flowers and leaves their distinctive shades of pink, purple, red, and blue.

They are categorised as secondary metabolites and produced by a particular flavonoid pathway branch. The antioxidant activity of anthocyanins is one of its key functions. They can scavenge free radicals and guard against oxidative damage to the plant cells. The plant's overall health and vitality are aided by its antioxidant capacity. Additionally, anthocyanins are essential for boosting tolerance to a range of biotic and abiotic stresses. They serve as a defence mechanism, preventing infections and herbivore attacks and minimising damage. Additionally, it has been discovered that anthocyanins can provide resistance to environmental challenges such as UV radiation, extreme heat, and drought. Genetic and environmental cues are just two examples of the many variables that influence how much anthocyanin a plant produces. Light, temperature, the availability of nutrients, and hormonal signals can all have an impact on how they express themselves. Anthocyanins have drawn a lot of interest for their possible health advantages for people in addition to their physiological roles in plants. They have been linked to a variety of beneficial impacts on human health, including anti-inflammatory and anti-cancer capabilities, and are acknowledged as powerful antioxidants. Foods high in anthocyanins have been related to better cardiovascular health, increased cognitive function, and a lower chance of developing chronic diseases.

Anthocyanins are significant plant metabolites that play crucial functions in both plant defence mechanisms and human health. They not only contribute to the vivid colours of flowers and leaves.[16]

2.2.4 ANTHRAQUINONES

The tricyclic aromatic compound family known as anthraquinones has the formula $C_{14}H_{10}O_2$. They are distinguished by the presence of ketone groups at the core ring's C-9 and C-10 locations. Because of their many substituent groups, anthraquinones can have a wide range of characteristics and uses. The term "hydroxyanthraquinoid derivatives" describes anthraquinone molecules in which hydroxyl (-OH) groups have been added in place of several hydrogen atoms. These derivatives' vibrant colours come from their pigmentation and capacity to absorb visible light. In many different industries, including food, cosmetics, and textiles, anthraquinones, notably hydroxyanthraquinoid derivatives, have been extensively used as natural colourants. Anthraquinones, which are employed as dyeing agents, are frequently found in *Rubia* species, also known as madder plants. Anthraquinones have exhibited a variety of

biological roles in addition to their colourant qualities, making them useful to the pharmaceutical business. A few anthraquinones have therapeutic effects, including antibacterial, antioxidant, anti-inflammatory, and anticancer characteristics. These substances are used in the creation of pharmaceutical medications and have had their potential therapeutic applications studied.[17]

Anthraquinones are significant substances in several industries, including colours and pharmaceuticals, thanks to their wide range of structural variations and biological activities. They are widely used and being investigated for possible uses because of their natural occurrence and advantageous characteristics.

2.2.6 TANNINS

Tannins have high molecular weights, ranging from 500 Da to more than 3000 Da, and are phenolic chemicals. Involvement in defence mechanisms against mammalian herbivores, birds, and insects is one of tannins' key roles in plants. They serve as deterrents by rendering the plant tissues poisonous or unpleasant to potential grazers. Tannins can assist in defending plants against microbial diseases. Due to the presence of several phenolic groups in their oligomeric structures, tannins can form complexes with proteins, carbohydrates, cellulose, and minerals. Their capacity to combine with other substances plays a role in the many biological processes and interactions that take place within the plant. Both flowering and non-flowering plants in the plant kingdom contain tannins. Numerous plant species are known to contain tannins, including the *Acacia* spp., *Sericea lespedeza*, and *Lotus* spp. These tannic-rich plants can be found all over the natural world. It can be difficult to define tannins precisely because they are a broad set of substances largely distinguished by their capacity to interact with proteins. Contrary to closely related phenolic compounds, which can be categorised according to their chemical makeup, tannins are categorised according to how water-soluble they are and whether they can form soluble or insoluble tannin-protein complexes.

Tannins have been investigated for their effects on protein metabolism in ruminant animals. They can lessen the amount of dietary protein degraded in the rumen, improving the way ruminants digest protein. Tannins may also improve an animal's ability to absorb amino acids in the small intestine, increasing its access to protein.

2.3 USE OF PHYTOCHEMICALS FOR THE TREATMENT OF LETHAL DISEASES

Plants have, in fact, been crucial to traditional medical procedures throughout history. Plants and their extracts have been used for thousands of years in traditional medical systems like Ayurveda and Traditional Chinese Medicine (TCM) to treat a variety of illnesses. The evolution of these conventional medical systems was aided by the transmission of information regarding plant selection, collection techniques, and medicine production down the generations. Recent scientific studies have offered proof in favour of the therapeutic abilities of phytochemicals contained in plants. These bioactive substances have demonstrated considerable promise in the treatment of a number of illnesses, including cancer. The origin of or a derivation from natural ingredients can be found in over 50% of the licenced anticancer medicines created between 1940 and 2014. A major field of research in the pharmaceutical sector has been the development of novel medicines from natural sources, including plants. Plants include a variety of chemical substances that have a wide range of biological functions that can be used therapeutically. Natural products have acted as a source of lead compounds in drug discovery and have inspired the creation of new therapeutic candidates. The mechanisms of action of phytochemicals having anticancer activities have been well investigated. These substances have the ability to have cytotoxic effects on cancer cells, to slow the growth of tumours, to cause apoptosis (programmed cell death), and to obstruct a number of molecular processes involved in the initiation and advancement of cancer.[18]

Table2: Literature review of using known phytochemicals for the treatment of various categories of cancer.

Phytochemical	Type of cancer	Assessment of the effect	Reference
Berberine	Colon Cancer	The Recurrence is stopped	[18]
Curcumin	Breast Cancer	Survival is free from progression	[18]

Epigallocatechin	Colon Cancer	Methylation pattern modification	[18]
Apigenin	Breast Cancer, Leukaemia	Modulate angiogenesis and apoptosis	[19]

The Phytochemicals obtained from plant sources have shown to be effective in treatment of various other diseases as well by acting as efficient protein or gene target causing inhibition of expression and preventing the spread of disease via various ways including Free Radicle scavenging, immune support, antimicrobial activity as well as anti-inflammatory activity which can help in treating hepatotoxicity, neurotoxicity, cardiovascular complications as well as digestive ailments.

CHAPTER 3: EXPERIMENTAL

3.1 CHEMICAL USED -

Conc. Sulphuric acid, Dilute ammonium hydroxide, Alcoholic Potassium hydroxide, Wagner reagent, Chloroform, Hydrochloric acid, Lead acetate, Ferric chloride, distilled water.

3.2 EQUIPMENTS –

Test tubes, conical flask, beakers, Whatman filter paper No 42, pipette, measuring cylinder, hot plate, weighing balance, Heating mantle, mortar and pestle, cultural tubes, test tube stand, centrifuge, Tissue paper, glass rod, falcon tubes, China dish, aluminium foil, etc.

3.3 PLANT MATERIALS -

Flowers of *Viola tricolor var. hortensis*, *Gazania rigens*, *Alcea Rosea*, *Dahlia pinnata*, *Chrysanthemum morifolium*.

3.4 METHODOLOGY

3.4.1 Collection of plant material

Flowers from a variety of plant species, including *Viola tricolour* var. *hortensis*, *Gazania rigens*, *Alcea Rosea*, *Dahlia pinnata*, and *Chrysanthemum morifolium*, were collected at Delhi Technological University. During the month of January, this collection took place. Only strong, healthy plants were chosen specifically for flower harvesting. The flowers were carefully selected and thoroughly washed to remove any latex or dirt that might have been present in order to maintain cleanliness.

3.4.2 Drying

Shade Dried –

The chosen plants (*Viola tricolour* var. *hortensis*, *Gazania rigens*, *Alcea Rosea*, *Dahlia pinnata*, and *Chrysanthemum morifolium*) were harvested, and the flowers were then thoroughly cleaned to remove any debris. The flowers were washed in two steps: first, using tap water, and then, to assure cleanliness, they were rinsed with double-distilled water.

The flowers were carefully distributed evenly between sheets of newsprint after being washed. After that, they were allowed to naturally dry for 15 days in the shade. In Delhi Technological University's plant biotechnology lab, this drying procedure was carried out at room temperature.

The dried flowers were crushed and finely powdered in a pestle and mortar when the allotted time had passed. The flowers were ground into a powder during this process.

Sun Dried –

After collection of all the flower samples from the different sources (*Viola tricolour* var. *hortensis*, *Gazania rigens*, *Alcea Rosea*, *Dahlia pinnata*, and *Chrysanthemum morifolium*). The samples are washed twice once with tap water and then again with distilled water. The flowers were then spread out on a newspaper and then left to dry under the sun for around 15 days. A fine powder was created by grinding using mortal & pestle as well as grinder. The resulting powder was prepared for further testing and analysis by being carefully put to a China dish and covered with aluminium foil.

3.4.3 Extract Preparation

Fresh Sample –

The petioles of *Chrysanthemum morifolium*, *Gazania rigens*, *Alcea rosea*, *Dahlia pinnata*, and *Viola tricolour* var. *hortensis* were cut off before the flowers were gathered. The flowers received two rinses with sterilised milli-Q water to remove any dust particles after being washed with running tap water to assure cleanliness. The flowers were ground into a fine paste using a mixer grinder to ensure consistency. In a 200 ml flask, 1 gram of the flower paste was combined with 100 ml of milli-Q water to create the plant extract. After boiling for 5 minutes, the liquid was then heated on a heating mantle for 10 minutes at a temperature of 50 C. The flask's contents were transferred to a 50 ml falcon tube after they had cooled to room temperature. The extract was centrifuged at 3000 rpm for five minutes to separate the leaf pellets from the supernatant. The supernatant was filtered through a Whatman filter paper, producing an aqueous extract of the flowers. The aqueous extract was then preserved for additional phytochemical analysis, enabling future research and tests.

Shade dried –

One gram of leaf powder was measured and put to a 200 ml flask with 100 ml of milli-Q water to create the aqueous extract of the leaves. The combination was then heated to a temperature of 50 C by placing the flask on a heating mantle. To make it easier to extract the necessary chemicals from the leaves, it was cooked for 5 minutes. The flask was allowed to cool naturally after boiling until it reached room temperature. The liquid was carefully transferred from the flask to a 50 ml falcon tube after it had cooled. The falcon tube containing the extract was centrifuged at a speed of 3000 rpm for 5 minutes to separate the liquid extract from the solid leaf particles. The supernatant was removed from the leaf pellets by running it through a Whatman filter paper in order to get a clean and filtered aqueous extract. Only the liquid extract was able to pass through and be collected because the filter paper had successfully caught the solid particles. The ensuing aqueous extract made from the leaves is represented by the collected supernatant, which can be further examined or put to use for a variety of uses.

Sun dried –

To a 200 ml flask, add 1 gram of leaf powder after measuring it out. As you carefully mix the powder and water in the flask, add 100 ml of milli-Q water. Heat the mixture in the flask to 50 C by setting it on a heating mantle. Give the mixture five minutes to

boil. The flask should be removed and left to naturally cool at room temperature. Make sure all of the mixture is transferred to a 50 ml falcon tube when the mixture has cooled. Centrifugate the falcon tube containing the extract for 5 minutes at 3000 rpm. Separate the supernatant from the remaining leaf matter before carefully pouring it from the falcon tube. Remove any leftover solid particles from the supernatant by filtering it through a Whatman filter paper. The subsequent aqueous extract made from the leaf is represented by the liquid that was collected, known as the supernatant. This aqueous extract may be subjected to additional examination, put to use in a variety of ways, or kept for later use.

2.5 QUALITATIVE ESTIMATION OF PHYTOCHEMICALS

- 3.5.1 Flavonoids Test

Each type of Flower extract was taken and were added in separate test tubes. For this test, 1 ml of each extract was to different test tubes. 10% lead acetate (1g Lead acetate in 10 ml water) solution was added to each test tube. The resulting mixture was observed to determine the reaction or outcome of the test.

- 3.5.2 Terpenoids Test

In a test tube 3 ml of extract was taken, 1ml of chloroform was added first and is followed by addition of 1.5 ml of concentrated H₂SO₄ along the sides of the test tube

- 3.5.3 Alkaloids Test

The qualitative test for alkaloids is Wagner's Test.

Wagner's reagent is prepared by adding 2.25 grams of iodine and 1.25 grams of potassium iodide to 250 ml of milli-Q water. After which 1 ml of the prepared plant extract was pipetted in a labelled test tube, Wagner reagent is added to it, approx. 1 ml

- 3.5.4 Anthocyanin Test

For the qualitative test of anthocyanin, to 2ml of extract 2N HCL was added to the test tube followed by the addition of diluted ammonia.

- 3.5.5 Anthraquinone Test

5 ml Extract was taken in a test tube and to this test tube a few ml of Conc. H₂SO₄ was added and stirred to this mixture diluted ammonia is added (1ml).

- 3.5.6 Tannins Test

Samples of all the different types were taken and 5 ml of each of the sample added in different tubes and were labelled accordingly. Some drops of neutral 5% ferric chloride to each test tube was added.

Table 3. Protocol for Qualitative test

S.no.	Preliminary Test	Procedure	Expected Results
1	Flavonoids	Extract + 10% lead acetate	Yellow Precipitate
2	Terpenoids	3 ml Extract + 1 ml Chloroform + 1.5 ml of concentrated H ₂ SO ₄	Reddish Brown color
3	Alkaloids	Extract + Wagner's Reagent	Reddish brown
4	Anthocyanins	2 ml extract + 2N HCL + Dil. Ammonia	Pink-red turns blue-violet
5	Anthraquinones	5ml of extract + few ml Conc. H ₂ SO ₄ + 1ml Dil. Ammonia	Rose pink colour
6	Tannins	5ml extract + 5% ferric chloride solution	Dark green color

2.6 THE TOOLS USED FOR IN SILICO ANALYSIS

In-silico analysis refers to the application of computational technologies to artificially simulate naturally occurring biological mechanism and make predictions based of the results obtained. In silico methods may be used to analyse certain genes that are linked

to a particular characteristic, illness, or biological function in order to predict targeted genes.

The tools that are used in this study are-

- **3.6.1 Pubchem**

It was used for obtaining the SMILE structure of the phytochemical that is being analysed

- **3.6.2 SwissTargetPrediction (([SwissTargetPrediction](#)))**

The SMILES structure that were obtained were fed into this software which gave results as prediction of the targets for the phytochemicals in question which can be further analysed and compared to find out the predictability of these molecules being an effective treatment method for certain lethal diseases.





CHAPTER 4: RESULTS

4.1 Qualitative estimation of Phytochemicals in *Alcea Rosea* for fresh, sun and shade dried Flowers

Table 4: Results showing the presence or absence of phytochemicals in *Alcea Rosea*

S. No.	Phytochemical	Fresh	Shade-Dried	Sun Dried
1	Flavonoids	-	-	-
2	Terpenoids	+	+	+
3	Alkaloids	+	+	+
4	Anthocyanins	-	-	-
5	Anthraquinones	+	+	+
6	Tannins	-	+	+

Table 5: Images of the results obtained in the 3 samples of fresh, shade and sun-dried flowers via different tests in *Alcea Rosea*.

Phytochemicals of <i>Alcea Rosea</i>	Results of the Fresh, Shade-dried & Sun-dried extracts
Flavonoids	
Terpenoids	
Alkaloids	
Anthocyanins	









- **The Phytochemicals that were found to be present in the Hollyhock sample are-**
Terpenoids, Alkaloids, Anthraquinones, Tannins
- **The Phytochemicals that were found to be absent in the Hollyhock sample are-**
Flavonoids, Anthocyanin

4.2 Qualitative estimation of Phytochemicals in *Viola tricolor var. hortensis*, for fresh, sun and shade dried Flowers

Table 6: Results showing the presence or absence of phytochemicals in *Viola tricolor var. hortensis*

S. No.	Phytochemical	Fresh	Shade-Dried	Sun Dried
1	Flavonoids	+	+	+
2	Terpenoids	-	+	+
3	Alkaloids	+	+	+
4	Anthocyanins	-	-	-
5	Anthraquinones	-	-	-
6	Tannins	-	+	+

Table 7: Images of the results obtained in the 3 samples of fresh, shade and sun-dried flowers via different tests in *Viola tricolor var. hortensis*.

Phytochemicals of <i>Viola tricolor var. hortensis</i>	Results of the Fresh, Shade-dried & Sun-dried extracts
Flavonoids	
Terpenoids	
Alkaloids	
Anthocyanins	
Anthraquinones	
Tannins	

- The Phytochemicals that were found to be present in the Pansy sample are-

Terpenoids, Alkaloids, Tannins, Flavonoids


- **The Phytochemicals that were found to be absent in the Pansy sample are-**
Anthraquinones, Anthocyanin






4.3 Qualitative estimation of Phytochemicals in *Dahlia pinnata* for fresh, sun and shade dried Flowers

Table 8: Results showing the presence or absence of phytochemicals in *Dahlia pinnata*

S. No.	Phytochemical	Fresh	Shade-Dried	Sun Dried
1	Flavonoids	+	+	+
2	Terpenoids	+	+	+
3	Alkaloids	+	+	+
4	Anthocyanins	-	-	+
5	Anthraquinones	+	+	+
6	Tannins	+	+	+

Table 9: Images of the results obtained in the 3 samples of fresh, shade and sun-dried flowers via different tests in *Dahlia pinnata*.

Phytochemicals of <i>Dahlia pinnata</i>	Results of the Fresh, Shade-dried & Sun-dried extracts
Flavonoids	

Terpenoids	
Alkaloids	
Anthocyanins	
Anthraquinones	
Tannins	




- **The Phytochemicals that were found to be present in the Dahlia sample are-** Terpenoids, Alkaloids, Tannins, Flavonoids, Anthraquinones,
- **The Phytochemicals that were found to be absent in the Dahlia sample are-** Anthocyanin (but was found in slight amounts in the sun-dried sample)




4.4 Qualitative estimation of Phytochemicals in *Gazania rigens* for fresh, sun and shade dried Flowers

Table 10: Results showing the presence or absence of phytochemicals in *Gazania rigens*

S. No.	Phytochemical	Fresh	Shade-Dried	Sun Dried
1	Flavonoids	-	-	-
2	Terpenoids	-	+	+
3	Alkaloids	+	+	+
4	Anthocyanins	-	-	-
5	Anthraquinones	-	-	-
6	Tannins	-	-	-

Table 11: Images of the results obtained in the 3 samples of fresh, shade and sun-dried flowers via different tests in *Gazania rigens*.

Phytochemicals of <i>Gazania rigens</i>	Results of the Fresh, Shade-dried & Sun-dried extracts
Flavonoids	
Terpenoids	
Alkaloids	

Anthocyanins	
Anthraquinones	
Tannins	

- The Phytochemicals that were found to be present in the Gazania sample are- Terpenoids (except the fresh sample), Alkaloids
- The Phytochemicals that were found to be absent in the Gazania sample are- Anthocyanin, Tannins, Flavonoids, Anthraquinones

4.5 Qualitative estimation of Phytochemicals in *Chrysanthemum morifolium* for fresh, sun and shade dried Flowers

Table 12: Results showing the presence or absence of phytochemicals in *Chrysanthemum morifolium*

S. No.	Phytochemical	Fresh	Shade-Dried	Sun Dried
1	Flavonoids	+	+	+
2	Terpenoids	-	+	-
3	Alkaloids	+	+	+
4	Anthocyanins	-	-	-
5	Anthraquinones	-	-	-

6	Tannins	+	+	+
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Phytochemicals of <i>Chrysanthemum morifolium</i>	Results of the Fresh, Shade-dried & Sun-dried extracts
Flavonoids	 <p>26/4/23 Flavonoid Shed CuSO₄</p> <p>26/4/23 Flavonoid Sun CuSO₄</p> <p>26/4/23 Flavonoid Fresh CuSO₄</p>
Terpenoids	 <p>26/4/23 Terpenoid Shed CuSO₄</p> <p>26/4/23 Terpenoid Sun CuSO₄</p> <p>26/4/23 Terpenoid Fresh CuSO₄</p>
Alkaloids	 <p>26/4/23 Alkaloid Fresh CuSO₄</p> <p>26/4/23 Alkaloid Shed CuSO₄</p> <p>26/4/23 Alkaloid Sun CuSO₄</p>
Anthocyanins	 <p>26/4/23 Anthocyanin Shed CuSO₄</p> <p>26/4/23 Anthocyanin Sun CuSO₄</p> <p>26/4/23 Anthocyanin Fresh CuSO₄</p>
Anthraquinones	 <p>26/4/23 Anthraquinone Shed CuSO₄</p> <p>26/4/23 Anthraquinone Sun CuSO₄</p> <p>26/4/23 Anthraquinone Fresh CuSO₄</p>
Tannins	 <p>26/4/23 Tannin Shed CuSO₄</p> <p>26/4/23 Tannin Sun CuSO₄</p> <p>26/4/23 Tannin Fresh CuSO₄</p>

Table 13: Images of the results obtained in the 3 samples of fresh, shade and sun-dried flowers via different tests in *Chrysanthemum morifolium*.

- **The Phytochemicals that were found to be present in the Gazania sample are-**
Terpenoids (except the fresh sample), Alkaloids, Tannins, Flavonoids
- **The Phytochemicals that were found to be absent in the Gazania sample are-**
Anthocyanin, Anthraquinones

4.6 TARGET PREDICTION FOR PHYTOCHEMICALS AND ANALYSIS

4.6.1 PHYTOCHEMICAL – APIGENIN

A flavonoid compound known as apigenin belongs to the structural class of flavones. 4',5,7-trihydroxyflavone is its scientific name, and its chemical formula is C₁₅H₁₀O₅. Apigenin has a low molecular weight of 270.24 and forms crystals that resemble yellow needles when it is pure. It melts at a temperature of 347.5 degrees Celsius. Apigenin does not readily dissolve in water and is nearly insoluble in it. In heated alcohol, it is just slightly soluble. Additionally, dimethyl sulfoxide (DMSO) and diluted potassium hydroxide can both dissolve it. The extraction of apigenin and its use in different applications both depend on its solubility qualities.[20]

Target	Common name	Uniprot ID	ChEMBL ID	Target Class	Probability*
NADPH oxidase 4	NOX4	Q9NPH5	CHEMBL1250375	Enzyme	1.0
Aldose reductase (<i>by homology</i>)	AKR1B1	P15121	CHEMBL1900	Enzyme	1.0
Cyclin-dependent kinase 5/CDK5 activator 1	CDK5R1 CDK5	Q15078 Q00535	CHEMBL1907600	Kinase	1.0
Xanthine dehydrogenase	XDH	P47989	CHEMBL1929	Oxidoreductase	1.0
Monoamine oxidase A	MAOA	P21397	CHEMBL1951	Oxidoreductase	1.0
Tyrosine-protein kinase receptor FLT3	FLT3	P36888	CHEMBL1974	Kinase	1.0
Cytochrome P450 19A1	CYP19A1	P11511	CHEMBL1978	Cytochrome P450	1.0
Estrogen receptor alpha	ESR1	P03372	CHEMBL206	Nuclear receptor	1.0
Cyclin-dependent kinase 1/cyclin B	CCNB3 CDK1 CCNB1 CCNB2	Q8WWL7 P06493 P14635 Q95067	CHEMBL2094127	Other cytosolic protein	1.0
Acetylcholinesterase	ACHE	P22303	CHEMBL220	Hydrolase	1.0
Adenosine A1 receptor (<i>by homology</i>)	ADORA1	P30542	CHEMBL226	Family A G protein-coupled receptor	1.0
Cyclooxygenase-2	PTGS2	P35354	CHEMBL230	Oxidoreductase	1.0
Estrogen receptor beta	ESR2	Q92731	CHEMBL242	Nuclear receptor	1.0
Cyclin-dependent kinase 6	CDK6	Q00534	CHEMBL2508	Kinase	1.0
Adenosine A2a receptor (<i>by homology</i>)	ADORA2A	P29274	CHEMBL251	Family A G protein-coupled receptor	1.0

Fig 7. Gene/Protein target prediction results for Apigenin

From Figure 7, Following points can be inferred -

- **NADPH Oxidase 4**, NOX4 shows a strong probability of being a gene target for apigenin which shows that it can be useful for the treatment of **pancreatic cancer**, **cardiac disorders like cardiac fibrosis** which can lead to **myocardial infarction** as well as **Lung disorders** as research has shown that NOX4 plays a crucial role in fibrosis of various organisms and The Akt-mTOR pathway can be activated by NOX4 to enhance protein synthesis, cell proliferation, and fibroblast activation, which can result in the formation of cardiac fibrosis.[21]
- **Cytochrome P450**, Strong probability is seen for CYP 450 family to be an efficient gene target for apigenin. From this it can be inferred that Apigenin can be an effective method of treatment for various varieties of cancers like Breast cancer, Hepatic cancer, endometrial cancer, Prostate cancer etc.[22]
- **Cyclin-dependent Kinase 6**, CDK-6 is found to have high chances of being an effective gene target for apigenin, which further shows that it can be used as an effective method for treatment of cancer as it has been shown that for certain variety of tumour cells CDK6 are crucial for cell proliferation.[23]

4.6.2 PHYTOCHEMICAL – QUERCETIN

A flavonoid substance called quercetin is naturally present in many different fruits, vegetables, cereals, and plants. It is categorised as a flavonol, a subclass of flavonoids renowned for its anti-inflammatory and antioxidant characteristics. Given that it was initially separated from the oak tree's bark, the name "quercetin" is taken from the Latin word "Quercetum," which meaning Oak Forest. Chemically speaking, quercetin is made up of a flavone backbone with hydroxyl groups connected in various places. Its chemical name, 3,3',4',5,7-pentahydroxyflavone, derives from its molecular formula, C₁₅H₁₀O₇. Due to its possible health benefits, quercetin has attracted a lot of interest in both science and medicine. It displays a variety of biological behaviours. It has shown cardioprotective, anti-viral, anticancer as well as anti-inflammatory activity.[24]

Target	Common name	Uniprot ID	ChEMBL ID	Target Class	Probability*
NADPH oxidase 4	NOX4	Q9NPH5	CHEMBL1250375	Enzyme	██████████
Vasopressin V2 receptor	AVPR2	P30518	CHEMBL1790	Family A G protein-coupled receptor	██████████
Aldose reductase	AKR1B1	P15121	CHEMBL1900	Enzyme	██████████
Xanthine dehydrogenase	XDH	P47989	CHEMBL1929	Oxidoreductase	██████████
Monoamine oxidase A	MAOA	P21397	CHEMBL1951	Oxidoreductase	██████████
Insulin-like growth factor I receptor	IGF1R	P08069	CHEMBL1957	Kinase	██████████
Tyrosine-protein kinase receptor FLT3	FLT3	P36888	CHEMBL1974	Kinase	██████████
Cytochrome P450 19A1	CYP19A1	P11511	CHEMBL1978	Cytochrome P450	██████████
Epidermal growth factor receptor erbB1	EGFR	P00533	CHEMBL203	Kinase	██████████
Thrombin	F2	P00734	CHEMBL204	Protease	██████████
Carbonic anhydrase II	CA2	P00918	CHEMBL205	Lyase	██████████
Serine/threonine-protein kinase PIM1	PIM1	P11309	CHEMBL2147	Kinase	██████████
Arachidonate 5-lipoxygenase	ALOX5	P09917	CHEMBL215	Oxidoreductase	██████████
Serine/threonine-protein kinase Aurora-B	AURKB	Q96GD4	CHEMBL2185	Kinase	██████████
Dopamine D4 receptor	DRD4	P21917	CHEMBL219	Family A G protein-coupled receptor	██████████

Fig 8. Gene/Protein target prediction results for Quercetin

From Figure 8, Following points can be inferred –

- **Vasopressin V2 receptor**, is predicted to be an efficient target for Quercetin, which implies that Quercetin can be effective in treatment of various renal complications like renal carcinoma and renal failure as the V2 receptor is found to play a crucial role in regulating the homeostasis of salt and water that is performed by the kidneys and it also is seen to stimulate the growth of the cancerous cell of the renal carcinoma.[25]
- **Thrombin**, is predicted to be an effective target for quercetin which implies that quercetin can be used to target and knock out thrombin thus preventing various cardiac complications like myocardial infarction as well as venous thromboembolism.[26]

4.6.3 PHYTOCHEMICAL – ISORHAMNETIN

The pharmacological effects of the flavonoid molecule isorhamnetin on a number of disorders have been investigated. Some of the impacts that have been mentioned are, Isorhamnetin has demonstrated potential cardioprotective effects by lowering oxidative stress, inflammation, and enhancing endothelial function. Cardiovascular disorders. It might aid in the management and prevention of cardiovascular disorders. Isorhamnetin has been researched for its possible anticancer effects, including tumour growth inhibition, apoptosis induction, and angiogenesis suppression.[27] It might be

therapeutically useful in the management of several tumour types. Neurodegenerative diseases: Research indicates that isorhamnetin has neuroprotective properties and might help to stop neurodegenerative conditions like Alzheimer's disease.

Inflammation, oxidative stress, and neural cell protection may all be decreased by it. Isorhamnetin's pharmacological effects are thought to be mediated by the control of a number of signalling pathways, including NF- κ B, PI3K/AKT, MAPK, and others. These pathways are crucial for biological activities like inflammation, cell proliferation, and cell survival.[27]

Target	Common name	Uniprot ID	ChEMBL ID	Target Class	Probability*
Xanthine dehydrogenase	XDH	P47989	CHEMBL1929	Oxidoreductase	
Carbonic anhydrase II	CA2	P00918	CHEMBL205	Lyase	
Carbonic anhydrase VII	CA7	P43166	CHEMBL2326	Lyase	
Carbonic anhydrase XII	CA12	O43570	CHEMBL3242	Lyase	
Carbonic anhydrase IV	CA4	P22748	CHEMBL3729	Lyase	
Cytochrome P450 1B1	CYP1B1	Q16678	CHEMBL4878	Cytochrome P450	
Multidrug resistance-associated protein 1	ABCC1	P33527	CHEMBL3004	Primary active transporter	
NADPH oxidase 4	NOX4	Q9NPH5	CHEMBL1250375	Enzyme	
Aldose reductase	AKR1B1	P15121	CHEMBL1900	Enzyme	
ATP-binding cassette sub-family G member 2	ABCG2	Q9UNQ0	CHEMBL5393	Primary active transporter	
Acetylcholinesterase	ACHE	P22303	CHEMBL220	Hydrolase	
Arachidonate 15-lipoxygenase	ALOX15	P16050	CHEMBL2903	Enzyme	
Arachidonate 12-lipoxygenase	ALOX12	P18054	CHEMBL3687	Enzyme	
Insulin-like growth factor I receptor	IGF1R	P08069	CHEMBL1957	Kinase	
Epidermal growth factor receptor erbB1	EGFR	P00533	CHEMBL203	Kinase	

Fig 9. Gene/Protein target prediction results for Isorhamnetin

From Figure 9, Following points can be inferred –

- **Carbonic anhydrase subclasses**, The subclasses of carbonic anhydrases like carbonic anhydrase XII are responsible for the regulation of pH, Metastasis and Cell adhesion, Targeting these subunits can help in prevention of cancer proliferation, and results here show that these subunits are effective targets for the Isorhamnetin compound.[28]

DISCUSSION

When Qualitative analysis was performed on the flowers of the five plant species, different phytochemicals like flavonoids, terpenoids, alkaloids, anthocyanins, anthraquinones and tannins were found to be present. These compounds have a large variety of role to play in the plant survival and also have numerous applications ranging from medical usage, cosmetics industry, Dye industry to supplements and many more. Alkaloids were found out to be present in the maximum number of samples which was followed by flavonoids and tannins. Anthocyanins and Anthraquinones were found in the least number of plant samples. These are bioactive compounds which offer a wide variety of medicinal usage as they are known to have various properties like anti-microbial activity, anti-inflammatory effect, Cardio-protective activity, hepatotoxicity prevention, neurotoxicity prevention, anti-oxidant property by free radicle scavenging, which allows them to be an effective treatment method for various types of diseases like different variety of cancers, Renal complications, Cardiovascular diseases like myocardial infarction, neurological disorders like Alzheimer's and Parkinson's Disease.

Additionally, In-silico analysis of the phytochemicals that were found in the greatest number of plant samples that were alkaloids and flavonoids lead to an understanding of the use of these phytochemicals as potential ligands for prevention of disease by the prediction of gene targets to which these compounds can bind to. Pubchem and SwissTargetPrediction facilitated this process of target prediction.

CONCLUSION

This Study shows the presence of different varieties of phytochemicals in the samples of the plants taken, which can be effectively used in various industries. The Qualitative analysis of the phytochemicals combined with the in-silico study performed help in the identification and understanding of the effectiveness of the phytochemicals as a method of treatment of various lethal diseases like breast cancer, lung cancer, Renal carcinoma, Myocardial infarction, Venous Thrombosis, Fibrosis, Hypertension, Parkinson's disease, digestive ailments, renal failure and many more. Use of Organically derived compounds can offer a safer alternative for treatment of these ailments as compared to the artificial or synthetic method which are prone to have a large number of side effects which can lead to a reduced quality of life. Use of phytochemicals for disease treatment offer a safer alternative and which can be cost effective as well as environmentally friendly as well. This study discussed about various secondary metabolites present in plants and their effectiveness for medicinal usage, Further research is needed to be performed in this field in order to obtain more concrete results and to have a better understanding of the application of phytochemicals as treatment methods. Randomized control trials can also help in better understanding the effectiveness of these novel treatment methods.

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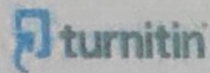
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