CHAPTER I
GENERAL INTRODUCTION

MAN’s adoration of mighty nature, the Eternal mother who blessed him with water to quench his thirst, soil to cultivate, plants for his food and fire to keep him warm, was prime of his feelings. As man is endowed with six senses and acquired skill, he began to exploit the inexhaustive resources of nature to improve the standard of his life. His adoration of nature has induced inquisitiveness [Ivanova et al., 2005].

The diverse hues of the sky reach the climax in the colourful rainbow whereas the wonderful garment of the Mother Earth possesses the complementary captivating part, the green vegetable kingdom. The plants serve as sources of timber, furniture, cotton, paper, resins, perfumes, spices, dyes, drugs etc. Man tried to distinguish the characteristics of various plants such as edible and nonedible, poisonous and nonpoisonous, medicinal and nonmedicinal plants and herbs [Ivanova et al., 2005].

Medicinal plants constitute the main source of new pharmaceuticals and health care products. The use of medicinal plants in the industrialized societies has been traced to the extraction and development of several drugs from these plants as well as from traditionally used folk medicine [Shrikumar and Ravi, 2007]. The secondary plant metabolites play important roles in human health and may be nutritionally important [Hertog et al., 1993]. Phytochemical screening of plants in various solvents has been revealed. Secondary metabolites of plants serve as defence mechanisms against predation by many microorganisms, insects and herbivores [Cowain, 1999]. Herbal medicines have become more popular in the treatment of many diseases due to popular belief that green medicine is safe, easily available with fewer side effects. According to the statistics furnished by INSD [Institut National de la Statistics et al la Demogra] more than 70% of people still rely on medicinal plants for the treatment of various diseases [INSD, 2007].

Phytochemical constituents and their uses

The valuable medicinal properties of different plants are due to the presence of several chemical constituents like alkaloids, tannins, phenolics, flavonoids,
terpenoids, carbohydrates, glycosides, steroids, saponins, fats and oils etc., Among them some are synergistic and enhance the bioactivity of other compounds.

Alkaloids

Alkaloids are defined as optically active, basic nitrogenous plant products containing nitrogen heterocycles in their structure. Alkaloid means alkali – like. Most of the alkaloids are derivatives of heterocyclic basic compound such as pyrrole, pyridine, quinoline and isoquinoline. Some alkaloids have open chain structure. Alkaloids are found in leaves, fruits, seeds, barks and roots of some dicotyledonous plants. They are abundant in the families like Apocynaceae, Papaveraceae, Rubiaceae, Rutaceae and Solanaceae. They are absent in lower plants like algae, fungi and gymnosperms [Evens and Trease, 1985].

They act as poisonous substances which afford plants safety from herbivores and insects. They act as reserve substances to supply nitrogen. They are the end products of detoxification mechanisms. They are considered as excretory products of plants and excess of ammonia is excreted. Some alkaloids inhibit enzyme activity.

Plant alkaloids are the primary active ingredients of Ayurvedic drugs. Alkaloid containing plants have been used by human since ancient times for the therapeutic and recreational purposes. Many alkaloids are still used in medicine. Morphine is a powerful narcotic used for relief of pain. Codeine, the methyl ether derivative of morphine found in the opium poppy, is an excellent analgesic that is relatively non addictive. Quinine, which is obtained from plants of the genus Cinchona, is used to treat arrhythmias of irregular heartbeat. Ergonovine is used to reduce uterine hemorrhage after child birth and ephedrine is used to relieve the discomfort of common colds, sinusitis, hay fever and bronchial asthma. Many alkaloids possess local anaesthetic properties, though clinically they are seldom used for this purpose. Cocaine is a very potent local anaesthetic. Quinine is a powerful antimalarial [Evens and Trease, 1985].

Tannins

Tannin (also known as vegetable tannin, natural organic tannins or sometimes tannoid, i.e., a type of biomolecule) is an astringent, bitter plant polyphenolic compound that binds to and precipitates proteins and various other
organic compounds including aminoacids and alkaloids. However, the term “tannin” by extension is widely applied to any large polyphenolic compound containing sufficient hydroxyls and other suitable groups (such as carboxyls) to form strong complexes with various macromolecules. The tannins are widely distributed in many species of plants, where they play a role in protection from predation and also as pesticides and in plant growth regulation [Katie et al., 2006]. They are heavily hydroxylated and polymerized to give the high molecular weight polyphenol. Typically, tannin molecules require atleast 12 hydroxyl groups and atleast 5 phenyl groups to function as protein binders. The tannin compounds can be classified into two major groups based on Goldbeater’s skin test. A group of tannins showing the positive tanning test may be regarded as true tannins while those, which are partly retained by the hide powder and fail to give the test, are called as Pseudotannins.

Tannins are medicinally significant due to their astringent properties. They promote rapid healing and the formation of new tissues on wounds and inflammed mucosa. Tannins are used in the treatment of varicose ulcers, hemorrhoids, minor burns, frostbite as well as inflammation of gums. Internally tannins are administered in cases of diarrhoea, intestinal catarrh and in cases of heavy metal poisoning as an antidote. Recently, these compounds have demonstrated their antiviral activities for treatment of viral diseases including AIDS [Vinod and Rangari, 2007]. They have anticarcinogenic and antimutagenic potentials which may be related to their antioxidative property, which is important in protecting cellular oxidative damage, including lipid peroxidation. The generation of superoxide radicals is reported to be inhibited by tannins. Tannins have also been reported to exert other physiological effects, such as to accelerate blood clotting, reduce blood pressure, decrease the serum lipid level, produce liver necrosis and modulate immune responses. The families of the plants rich in both of the above groups of tannins include Rosaceae, Geraniaceae, Leguminosae, Combretaceae, Rubiaceae, Polygonoaceae, Theaceae etc., In the plants in which tannins are present, they exert an inhibitory effect on many enzymes due to their nature of protein precipitation and therefore contribute a protective function in barks and heartwood [Evens and Trease, 1985].
Phenolics

Phenols, sometimes called phenolics, having a hydroxyl group (-OH) is bonded directly to an aromatic hydrocarbon group. Phenolic compounds are classified as simple phenols or polyphenols based on the number of phenol units in the molecule [Rebecca and Robbins, 2003]. Hundreds of natural phenol compounds have been identified from medicinal herbs and dietary plants, mainly including phenolic acids, flavonoids, tannins, stilbenes, lignans, quinines etc. Their physiological and pharmacological functions may originate from their antioxidant and free radical scavenging properties and function of regulating detoxifying enzymes. Further, these antioxidant activities are related to the structures of phenolic compounds, generally depending on the number and positions of hydroxyl groups and glycosylation or other substituent [Cal et al., 2006; Heim et al., 2002].

In the plants, the phenolic units are esterified or methylated and are subjected to conjugation, which means that the natural phenols are mostly found in the glycoside form instead of the aglycone form. Phenolic compounds act as protective agents, inhibitors, natural animal toxicants and pesticides against invading organisms such as herbivores, nematodes, pnutophagous insects and fungal and bacerisla pathogens. The scent and pigmentation conferred by other phenolics can attract symbiotic microbes, pollinators and animals [Bhattacharya, 2010].

Phenolic compounds are present in food consumed in human diets and in plants used in traditional medicine of several cultures. Their role in human health and disease is a subject of research. Some phenols are germicidal and are used in formulating disinfectants. Others possess estrogenic or endocrine disrupting activity [Khoddami, 2013; Klepacka, 2011; Mishra and Tiwari, 2011; Robert Wildman, 2006].

Carbohydrates

Carbohydrates are technically hydrates of carbon, which comprises only carbon, hydrogen and oxygen, usually with hydrogen: oxygen atom ratio of 2:1 (as in water); structurally it is more accurate to view them as polyhydroxy aldehydes and ketones. The saccharides are divided into four chemical groupings: monosaccharides, disaccharides, oligosaccharides and polysaccharides. In general,
the monosaccharides and disaccharides, which are smaller (lower molecular weight) carbohydrates, are commonly referred to as sugars [Flitsch et al., 2003].

Carbohydrates originate as product of photo synthesis, an endothermic reductive condensation of carbon dioxide requiring light energy and the pigment chlorophyll.

\[ n\text{CO}_2 + n\text{H}_2\text{O} \xrightarrow{\text{hv}} \text{C}_n\text{H}_{2n}\text{O}_n + n\text{O}_2 \]

Carbohydrates perform numerous roles in living organisms. Polysaccharides are major source of metabolic energy, both for plants and animals. The 5-carbon monosaccharide ribose is an important component of coenzyme (eg. ATP, FAD and NAD) and the backbone of the genetic molecule known as RNA. The related deoxyribose is a component of DNA. Saccharides and their derivatives play key roles in the immune system, fertilization, preventing pathogenesis, blood clotting and development [Maton Anthea et al., 1993].

Aminoacids / Proteins

Amino acids are biologically important organic compounds made from amine (-NH$_2$) and carboxylic acid (-COOH) functional groups, along with a side chain specific to each amino acid. About 500 amino acids are known. Structurally they can be classified according to the functional group locations as alpha (\(\alpha\)-), beta (\(\beta\)-), gamma (\(\gamma\)-) or delta (\(\delta\)-) amino acids. In the form of proteins, amino acids comprise the second largest component (after water) of human muscles, cells and other tissues [Wagner and Ingrid, 1983].

Many important proteinogenic and non-proteinogenic amino acids play critical non-protein roles within the body. For example, in the human brain, glytamate (standard glutamic acid) and gamma-amino-butryic acid (“GABA”, non-standard gamma-amino acid) are respectively the main excitatory and inhibitory neurotransmitters, hydroxyproline (a major component of the connective tissue collagen) is synthesized from proline; the standard amino acid glycine is used to synthesise porphyrins used in red blood cells and the non-standard carnitine is used in lipid transport [Petroff, 2002].
Nine of the twenty standard amino acids are “essential” for humans because they cannot be created from other compounds by the human body and so must be taken in as food. Other amino acids may be essential for certain ages and also for some medical conditions. Because of their biological significance, amino acids are important in nutrition and are commonly used in nutritional supplements, fertilizers and food technology [Petroff, 2002].

Terpenoids

The terpenoids (isoprenoids) are naturally occurring organic chemicals similar to terpenes. Terpenes are hydrocarbons resulting from the combination of several isoprene Units. The terpenoids can be classified on the basis of the number of isoprene units used: hemiterpenoids, monoterpenoids, sesquiterpenoids, diterpenoids, triterpenoids, tetraterpenoids and polyterpenoids. Terpenes are widespread in the plant kingdom. The odours of flowers, fruits, leaves and wood of several trees are due to terpenes. They are responsible for the characteristic smell of eucalyptus, rose, citrus, lemon grass etc., Vitamin A is a terpene and another terpene, carotene is the precursor of vitamin A.

There have been many applications of terpenes in human societies. Pharmaceutical and food industries have exploited them for their potentials and effectiveness as medicines and flavor enhancers. Perhaps the most widely known terpene is rubber, which has been used extensively by humans. Rubber is a polyterpene, composed of repeating subunits of isoprene. The addition of sulfur to rubber by Charles Goodyear led to vulcanized rubber, which yields various degrees of pliability depending on the mixture ratio [Stiehler and Wakelin, 1947]. Other important terpenes include camphor, menthol, pyrethrins (insecticides), cleaners, antiallergenic agents and solvents. Rosin (a diterpene), limonene, carvone, nepetalactone (in catnip), hecogenin (a detergent) and digitoxigenin are also important terpenes [Croteau et al., 2000].

Agriculture has also shown an increasing interest in terpenes. In a study by Villalba et al., (2006) sheep were suggested to have increased tolerance for terpene consumption if they consumed more grains. They also showed terpenes can influence ungulate herbivory on other plants. This may help agronomists balance
diets of ruminants if they consume plants such as sagebrush (Artemesia sp.). Terpenes have also shown antimicrobial activities [Islam et al., 2003]. This is important due to the increase in antibiotic resistant bacteria, which is occurring globally and at an alarming rate. Addition of terpenes into livestock feed may replace conventional antibiotic addition, which in turn would slow the rate of antibiotic resistance in bacteria. The effect of some terpenes on microorganisms has been seriously studied since at least the 1980's [Andrews et al., 1980]. Plant oils, which contain terpenes, have shown increasing promise in vivo, inhibiting multiple species of bacteria. For example, cinnamon oil has shown broad-spectrum activity against Pseudomonas aeruginosa [Prabuseenivasan et al., 2006]. The various compositions of terpenes can be markedly different from one species to another. For example, John et al., (2007) found plant oils from Neolitsea foliosa, which also showed some antibacterial properties, included sesquiterpenes such as caryophyllene but lacked monoterpenes.

To better understand the vast array of terpenes, genetically modified organisms have been used. For example, the biosynthesis of terpenes has been studied in transformed E. coli [Adam et al., 2002]. As described by Adam et al., (2002), modification of organisms is important to help understand the various pathways of terpene synthesis for the purpose of producing antimicrobial and antiparasitic drugs [Goulart et al., 2004].

Fats

Fats consist of a wide group of compounds that are generally soluble in organic solvents and insoluble in water. Chemically, fats are triglycerides: Triesters of glycerol and fatty acids. Fats may be either solid or liquid at room temperature, depending on their structure and composition. “Oils” are usually used to refer to fats that are liquids at room temperature, while “Fats” are usually used to refer to fats that are solids at room temperature. “Lipids” are used to refer to both liquid and solid fats, along with other related substances, usually in a medical or biochemical context, which are not soluble in water. Fats can be categorized into saturated fats and unsaturated fats. Unsaturated fats can be further classified into cis fats, which are the most common in nature and trans fats, which are rare in nature but present in
partially hydrogenated vegetable oils. Trans fats may significantly increase the risk of coronary heart diseases [Evens and Trease, 1985].

Fats are an important part of diet of most heterotrophs (including humans). Fats or lipids are broken down in the body by enzymes called lipases produced in the pancreas. Vitamin A, D, E and K are fat soluble, meaning they can only be digested, absorbed and transported in conjugation with fats. Fats are also sources of essential fatty acids and important dietary requirement. Fats play a vital role in maintaining healthy skin and hair, insulating body organs against shock, maintaining body temperature and promoting healthy cell functions. Fats also serve as energy stores for the body, containing about 37 kilojoules per gram. They are broken down in the body to release glycerol by the liver and thus used as a source of energy.

Fats also serve as a useful buffer towards a host of diseases. When a particular substance, whether chemical or biotic-reaches unsafe levels in the bloodstream, the body can effectively dilute or at least maintain equilibrium of the offending substances by storing it in new fat tissue. This helps to protect vital organs, until such time as the offending substances can be metabolized and/or removed from the body by such means as excretion, urination, accidental or intentional bloodletting, serum excretion and hair growth. While it is nearly impossible to remove fat completely from the diet, it would also be unhealthy to do so [Evens and Trease, 1985].

Saponins

Saponins are glucosides with foaming characteristics. Saponins consists of a polycyclic aglycones attached to one or more sugar side chains. The aglycone part, which is also called sapogenin, is either steroid (C27) or a triterpene (C30). The foaming ability of saponins is caused by the combination of a hydrophobic (fat-soluble) sapogenin and a hydrophilic (water-soluble) sugar part. Saponions have a bitter taste. Some saponins are toxic and are known as sapotoxin. Phytochemical saponins can be found in most Vegetables, beans and herbs. Commercial saponins are extracted mainly from Yucca schidigera and Quillaja saponaria [Evens and Trease, 1985].
Saponins have many health benefits. They cause a reduction of blood cholesterol by preventing its re-absorption. They have antitumor and anti-mutagenic activities and cause lowering the risk of human cancers, by preventing cancer cells from growing. When ingested by humans, saponins also seem to help our immune system and to protect against viruses and bacteria. The non-sugar parts of saponins have also a direct antioxidant activity, which may result in other benefits such as reduced risk of cancer and heart diseases [Markham, 1982].

Steroids

A steroid has a characteristic arrangement of four cycloalkane rings that are joined to each other. The dietary fat cholesterol, the sex hormones estradiol and testosterone and the anti-inflammatory drug dexamethasone are some examples. The core of steroids is composed of twenty carbon atoms bonded together that take the form of four fused rings: Three cyclohexane rings (designated as rings A, B and C) and one cyclopentane ring (the D ring). The steroids vary by the functional groups attached to this four-ring core and by the oxidation state of the rings. Sterols are special forms of steroids, with a hydroxyl group [Moss, 1989]. Hundreds of distinct steroids are found in plants, animals and fungi. All steroids are made either from the sterols lanosterol (animals and fungi) or from cycloartenol (plants). Both lanosterol and cycloartenol are derived from the cyclization of the triterpene squalene [“Lanosterol biosynthesis”-Recommendations on Biochemical and Organic Nomenclature, Symbols and Terminology, International Union of Biochemistry and Molecular Biology [Hanukoglu, 1992].

Steroidogenesis is the biological process by which steroids are generated from cholesterol and transformed into other steroids. The major classes of steroid hormones in the human steroidogenesis are progesterone, corticosteroids, androgens and estrogens. Progesterone serves as precursors to all other human steroids—thus all human tissues which produce steroids must first convert cholesterol to pregnenolono. This conversion is the rate limiting step of steroid synthesis, which occurs inside the mitochondria of the respective tissue [Rossier, 2006]. Corticosteroids are produced in the adrenal cortex. Estrogen and progesterone are made primarily in the ovary and in the placenta during pregnancy and testosterone in
the testes. Testosterone is also converted into estrogen to regulate the supply of each, in the bodies of both females and males. In addition, certain neurons and glia in the central nervous system (CNS) express the enzymes that are required for the local synthesis of pregnane neurosteroids, either de novo or from peripherally derived sources.

Flavonoids

The term flavonoid (Lat, Flavus = yellow) was coined by Geissman and Hinreiner [Geissman et al., 1975] Kostanecki and Tambor [Von Konstanecki, 1985] suggested the name flavone first.

Flavonoids are the low molecular weight, heterocyclic, polyphenolic, secondary metabolic compounds that occur naturally in plants [Hollman et al., 1997]. Different kinds of flavonoids like Chalcones, dihydrochalcones, flavonols, flavanols, flavan-3, 4 – diols, flavones, flavanones, anthocyanidines, iso flavones, iso flavanols, iso flavanones and aurones are present in all dietary plants and medicinal herbs. The presence of one or more of the flavonoids graces the plants. About 2% of the total carbon photosynthesized by plants is converted to flavonoids [Markham, 1982].

Flavonols are the most abundant flavonoids in foods, with quercetin, keampferol and myricetin being the three most common flavonoids. Flavanones are mainly found in citrus fruit and flavones in celery. Catachins are present in large amounts in green and black tea and in red wine, whereas anthocyanins are found in strawberries and other berries. Iso flavones are almost exclusively found in soy foods [Loic Le Marchand, 2002].

These compounds serve essential functions in plant reproduction by recruiting, pollinators and seed dispersers. They are also responsible for the beautiful colour in many plant species, which has recently been suggested to protect leaf cells from photo-oxidative damage, thereby enhancing the efficiency of nutrient retrieval during senescence [Field et al.,2001]. Flavonoids display a broad spectrum of bioactivities such as anticancer, antifungal, antibacterial, antiviral, anti-inflammatory and antioxidant properties.
Taxonomically the most valuable substances are those that are relatively stable as end products of metabolism. Of all secondary substances, by far the greatest diversity in structure is shown by those compounds that contain phenolic groups. Approximately 36% of the naturally occurring compounds of plants contain phenolic hydroxyls and of these about one – third are flavonoid types [Karrer, 1958].

A number of monographs are available providing voluminous information on the isolation, synthesis, biochemistry, biogenesis, stereochemistry, biological activity and economic importance of flavonoids [Mabry et al., 1970; Harborne, 1988; Heim et al., 2002; Patel, 2008].

Flavonoids form the largest single family of naturally occurring oxygen containing heterocyclic compounds. Benzo – γ – pyrone (I) of various levels of saturation, oxidation and substitution constitutes the basic skeleton for these polyphenolics. 4 – Chromanone (II) substituted by a phenyl ring at C – 2 forms flavanones (IIIa) and flavanonols (dihydroflavonols) (IIIb). Flavones (IVa) and flavonols (IVb) which are the co-pigments in both cyanic and acyanic flowers are formed by substitution of a phenyl ring at C – 2 position of the chromone nucleus. Also included in this category is the flavan – 3, 4 – diols (proanthocyanidins) (V), anthocyanidins (VI) which occur as scarlet, red, mauve and blue flower pigments.

![Chemical Structures]

I  Benzo – γ – pyrone (Chromone)  
II  4 – Chromanone
(a) $R^1, R^2, R^3, R^4, R^5, R^6 = H$, Flavanone
(b) $R^2, R^3, R^4, R^5, R^6 = H, R^1 = OH$, Flavanone

(c) $R^1, R^3, R^5 = H, R^2, R^4 = OH, R^6 = OCH_3, 5, 7 – dihydroxy – 4′ – methoxy flavanone$ (Isosakuranetin)

(d) $R^1, R^3 = H, R^2, R^5 = OH, R^4, R^6 = OCH_3, 5, 3′ – dihydroxy – 7,4′ – dimethoxy flavanone$ (Persicogenin)

(e) $R^1, R^5 = H, R^2, R^3, R^4, R^6 = OCH_3, 5, 6, 7, 4′ – tetramethoxy flavanone$

(f) $R^1, R^5 = H, R^6 = OH, R^2, R^3, R^4 = OCH_3, 4′$-hydroxy – 5, 6, 7 – trimethoxy flavanone

(g) $R^1, R^5 = H, R^2, R^3, R^4 = OH, R^6 = OCH_3, 5, 6, 7 – trihydroxy – 4′ – methoxy flavanone$

(a) $R^1, R^2, R^3, R^4, R^5, R^6 = H$, Flavone
(b) $R^2, R^3, R^4, R^5, R^6 = H, R^1 = OH$, Flavonol
(c) R¹ = H, R², R⁴, R⁶ = OH, R³, R⁵ = OCH₃, 5, 7, 4’ – trihydroxy – 3’, 6 – dimethoxy flavone (Jaceosidin)

(d) R¹ = H, R², R⁶ = OH, R³, R⁴, R⁵ = OCH₃, 5, 4’ – dihydroxy – 4’, 6, 7 – trimethoxy flavone (Cirsilineol)

(e) R¹ = H, R², R³ = OH, R⁴, R⁵, R⁶ = OCH₃, 5, 6 – dihydroxy – 3’, 4’, 7 – trimethoxy flavones (Eupatilin)

(f) R¹ = H, R² = OH, R³, R⁴, R⁵, R⁶ = OCH₃, 3’ – O – methyl Eupatilin

(g) R¹ = H, R², R⁴, R⁵, R⁶ = OH, R³ = OCH₃, 5, 7, 3’, 4’ – tetrahydroxy – 6 – methoxy flavones (Nepetin)

\[
\begin{align*}
\text{Flavan – 3, 4 – diol} \\
\end{align*}
\]

\[
\begin{align*}
\text{Anthocyanidin} \\
\end{align*}
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Leucoanthocyanidins (VII) which are found in the heartwoods and in the leaves of woody plants, glycoflavones which contain C – C linked sugars as is exemplified by vitexin (VIIIa) and biflavonyls that are formed by carbon – carbon or carbon – oxygen coupling between two flavone units as in amentoflavone (IX) or hinokiflavone (X) are confined to the gymnosperms. Chalcones (phenylstyrylketones) (XI), dihydrochalcones (3 – phenylpropiofenones) (XII),
isoflavones (3 – phenylchromones) (XIII), neoflavones (4 – phenylcoumarins) (XIVa) and aurones (Lat, aurum = gold) (2 – benzilidine – 3 – coumarones) (XV) do not actually possess the basic 2 – phenylchroman but are so closely related chemically and biosynthetically to the other flavonoid types.

All these heterocyclics are made up of two distinct units, the C₆ – C₃ fragment that includes the B – ring and the C₆ fragment, A – ring. The flavonoids originate from a C₆ – C₅ – (C₂ + C₂ + C₂) precursor, the last six carbon atoms being found in the aromatic A – ring. The carbon atoms are referred to by a numbering system which utilizes ordinary numerals for the A – and C – rings and primed numbers for the B – ring. However, a modified numbering is adopted in the case of chalcones.

![Diagram of flavonoids](image)

(a) R¹ = Glu, R² = R³ = H, Vitexin
(b) R¹ = R² = H, R³ = Glu, Isovitexin (Saponaretin)
(c) R¹ = H, R² = R³ = Glu, Saponarin
(d) R¹ = Xylo – Glu, R² = R³ = H, Xylosyl vitexin.
IX
Amentoflavone (Biflavone)

X
Hinokiflavone

XIa-Chalcone

XII-Chalcone
The various flavonoids differ from one another only in the state of oxidation of the central ring. The individual compounds within each class are distinguished mainly by the number and location of hydroxyl, methoxyl and other groups substituted on the two benzene rings. These groups are usually arranged in restricted patterns in the flavonoid molecules reflecting the different biosynthetic origin of the two aromatic rings. The hydroxylation pattern of the A – ring originates from the acetate or malonate unit while that of B – ring resembles that found in the commonly occurring cinnamic acids viz. p – coumaric acid (XVIa), caffeic acid (XVIb), ferulic acid (XVIc) and sinapic acid (XVID) and coumarins (XIVb) and reflects their common biosynthetic origin from prephenic acid (XVII) and its congeners [Wong, 1976]. In general, flavonoids are related biosynthetically to aromatic amino acids phenylalanine (XVIIIa) and tyrosine (XVIIIb) through the corresponding cinnamic acids and the basic C15 skeleton is probably derived in plants by the coupling of a C6 – C3 unit produced by shikimic acid (XIX) path way and three C2 acetate units [Harborne, 1973].
Flavonoids occur commonly as flavonoid O – glycosides in which one or more of the flavonoid hydroxyls are involved in linkage with C – 1 of a sugar through an acid – labile hemiacetal bond. In vivo the flavonoids largely exist as glycosides [Britton, 1983]. The sugar –free compounds are referred to as aglycones. Glycosylation not only confers sap solubility to the somewhat insoluble flavonoid aglycones but also confers stability especially for the highly hydroxylated compounds.

The ability of flavonoids to form glycosides with a number of rarer sugars viz. sugars that are not part of the common metabolism might be related to the transport of such saccharides in a non reactive form to site of synthesis of gums and mucilages in which they are found [Pridham, 1960]. Glycosylation also serves to protect the flavonoids themselves from attack by oxidizing enzymes [Baruah and Swain, 1959] and also their translocation in plants [Roberts et al., 1959].
Prephenic acid (a) \( R = H \), Phenylalanine
(b) \( R = \text{OH} \), Tyrosine
Shikimic acid

There is a considerable range of flavonol glycosides present in plants. Over seventy different glycosides of quercetin alone have been described [Harborne, 1984]. By far the commonest is quercetin 3 – O – rutinoside (rutin) (XXa). Flavones occur as glycoside but the range of different glycosides is less than those observed in the case of flavonols. Apigenin 7 – O – \( \beta \) – glucopyranoside (XXIa) is a typical example of flavone glycoside.

Hydroxyls in certain sites have higher probability for glycosylation viz. C – 7 hydroxyl in flavones and isoflavones, C – 3 and C – 7 hydroxyls in flavonols and dihydroflavonols and C – 3 and C – 5 hydroxyls in anthocyanidins. The sugars which have been found in flavonoid glycosides include simple hexoses and pentoses (monosides) or di – and tri – saccharides (biosides and triosides) always combined through the oxygen at the C – 1 (anomeric centre) usually through a \( \beta \) – link; it is \( \alpha \) – only in the case of rhamnoses and arabinoses. Oligosaccharides of flavonoids are generally restricted to those with 1\( \rightarrow \)2 and 1\( \rightarrow \)6 linkages and they usually have glucose at the reducing end. A tetraglycoside viz. kaempferol 3 – O – sophorotrioside – 7 – O – rhamnoside (XXIIa) is reported from the seeds of Solanum tuberosum [Harborne, 1967] and wild potato [Harborne, 1962].
(a) $R^1 = \text{Rut, } R^2, R^3, R^4, R^5 = H$, Quercetin 3 – O – rutinoside (rutin)
(b) $R^1, R^2, R^3, R^4, R^5 = H$, Quercetin
(c) $R^1 = \text{Rha, } R^2, R^3, R^4, R^5 = H$, Quercetin 3 – O – rhamnoside (quercitrin)
(d) $R^1, R^2, R^3, R^4, R^5 = \text{Me}$, Quercetin pentamethyl ether
(e) $R^1 = \text{Glu, } R^2, R^3, R^4, R^5 = H$, Quercetin 3 – O – glucoside
(f) $R^1, R^3, R^5 = \text{Me, } R^2, R^4 = H$, Quercetin 3, 4', 7 – trimethyl ether (ayanin)
(g) $R^1 = \text{Gal, } R^2, R^3, R^4, R^5 = H$, Quercetin 3 – O – galactoside (hyperoside)
(h) $R^1, R^2, R^4, R^5 = H, R^3 = \text{Glu}$, Quercetin 7-O-glucoside (quercimeritrin)
(i) $R^1 = \text{Xyl – Gal, } R^2, R^3, R^4, R^5 = H$, Quercetin 3 – O – xylosylgalactoside
(j) $R^1 = \text{Xyl – Glu, } R^2, R^3, R^4, R^5 = H$, Quercetin 3-O-sambubioside
(k) $R^1, R^2 = H, R^3, R^4 = \text{Me, } R^5 = \text{Glu}$, Rhamnazin 4' – O – glucoside
(l) $R^1, R^2, R^4, R^5 = H, R^3 = \text{Xyl}$, Quercetin 7 – O – xyloside
(m) $R^1 = \text{Glu – Ara, } R^2, R^3, R^4, R^5 = H$, Quercetin 3 – O – glucoarabinoside
(n) $R^1 = \text{Me, } R^2, R^3, R^4, R^5 = H$, Quercetin 3 – methyl ether
(o) $R^1 = \text{Me, } R^5 = \text{Glu, } R^2, R^3, R^4 = H$, Quercetin 3 – methyl ether 4' – O – glucoside
(p) $R^1 = \text{Gal – Glu, } R^2, R^3, R^4, R^5 = H$, Quercetin 3 – O – galactosylglucoside
(a) $R^2 = \beta - \text{Gluco (pyranosyl)}, R^1, R^3, R^4 = H$, Apigenin $7 - O - \beta - \text{glucopyranoside}$

(b) $R^2, R^4 = \text{Glu}, R^1, R^3 = H$ Apigenin $4',7 - O$ diglucoside

(c) $R^2 = \text{Rha} - \text{Glu}, R^1, R^3, R^4 = H$, Apigenin $7 - O$ rhamnoglucoide

(d) $R^1, R^2, R^3, R^4 = H$, Apigenin

(e) $R^2 = \text{Glucur}, R^1, R^3, R^4 = H$, Apigenin $7 - O$ glucuronide

(f) $R^2 = 6'' - \text{ethylglucur}, R^1, R^3, R^4 = H$, Apigenin $7 - (6'' - \text{ethylglucuronide})$

(g) $R^2 = \text{Glucur}, R^4 = 6''$ malonylgluco, $R^1, R^3 = H$, Apigenin $4' - (6'' - \text{malonylglucoside}) 7 - \text{glucuronide}$

(h) $R^2 = \text{Gal}, R^3 = \text{Me}, R^1, R^3 = H$, Acacetin $7 - \text{galatoside}$. 

Glucose (XXIIIa), galactose (XXIVa), rhamnose (XXV), xylose (XXVI), arabinose (XXVII), apiose (XXVIII), glucuronic acid (XXIIIb) and galacturonic acid (XXIVb) are the monosaccharides and rutinose ($O - \alpha - L - \text{rhamnosyl (1} \rightarrow 6\text{) glucose}$) (XXIX), neohesperidose ($O - \alpha - L - \text{rhamnosyl (1} \rightarrow 2\text{) glucose}$) (XXX) and sophorose ($O - \beta - D - \text{glucosyl (1} \rightarrow 2\text{) glucose}$) (XXXI) are the disaccharides most commonly involved in glycosylation. Acetylated glycosides have also been encountered where one or more sugar hydroxyls are derivatised with an acid such as acetic or ferulic acid. Within a plant it has been observed that the glycosidic pattern may be relatively constant [Crowden et al., 1969; Harborne et al., 1972].
(a) $R^1 = \text{sopho}_3, R^2, R^4 = H, R^3 = \text{Rha}, \text{Kaempferol} 3 – O – \text{sophorotrioside – 7 – O – rhamnoside}$

(b) $R^1, R^2, R^3, R^4 = H, \text{Kaempferol}$

(c) $R^1 = \text{Ara}, R^2, R^3, R^4 = H, \text{kaempferol} 3 – O – \text{arabinoside}$

(d) $R^1 = \text{Glucur}, R^2, R^3 = H, R^4 = \text{Me}, \text{kaempferide} 3 – O – \text{glucuronide}$

(e) $R^1 = \text{Glu}, R^2, R^3, R^4 = H, \text{kaempferol} 3 – O – \text{glucoside}$

(f) $R^1 = \text{Rut}, R^2, R^3, R^4 = H, \text{kaempferol} 3 – O – \text{rutinoside}$

(g) $R^1 = \text{(sopho)}, R^2, R^3, R^4 = H, \text{kaempferol} 3 – O – \text{sophoroside}$

(h) $R^1 = \text{Neohesp}, R^2, R^3, R^4 = H, \text{kaempferol} 3 – O – \text{neohesperidoside}$

(i) $R^1, R^2 = \text{Gal}, R^3, R^4 = H, \text{kaempferol} 3, 5 – O – \text{digalactoside}$

(j) $R^1, R^2, R^4 = H, R^3 = \text{Glu}, \text{kaempferol} 7 – O – \text{glucoside (populnin)}$

(k) $R^1 = \text{Gal}, R^2, R^3, R^4 = H, \text{kaempferol} 3 – O – \text{galactoside}$

(l) $R^1, R^3 = \text{Glu}, R^2, R^4 = H, \text{kaempferol} 3, 7 – \text{di – O – glucoside}$

(m) $R^1 = \text{Me}, R^2, R^3, R^4 = H, \text{Isokaempferide}$.

The enthralling colours of the plants, flowers and fruits are due to the presence of one or more of the flavonoids. The flavonoids are certainly one among the sundry floral pigments [Harborne, 1967]. The flavonoid colours in flower petals are the means of attracting insects and animals and ensuring fertilization [Thompson et al., 1972]. The coloured anthocyanins in flower petals are almost invariably accompanied by flavones and flavonols. Flavones are important co – pigments, being essential for the full expression of anthocyanin colour in floral tissues [Asen et al., 1972] Gossypetin (XXXIIa), quercetagetin (XXXIII), syringetin (XXXIVa) and
isorhamnetin (XXXVa) are some of the few flavonols which are pigments in their own right providing yellow flower colours. Flavonols contribute to yellow colour particularly when they are methylated or when they are present in an unusual glycosidic form [Goodwin, 1976].

(a) R = CH₂OH, β – D – Glucose

(b) R = COOH, D – Glucuronic acid

(a) R = CH₂OH, β – D – Galactose

(b) R = COOH, D – Galacturonic acid
\[ \alpha - L - \text{Rhamnose} \]

\[ \beta - D - \text{Xylose} \]

\[ \alpha - L - \text{Arabinose} \]

\[ D - \text{Apiose} \]
Rutinose \([\alpha - L - \text{Rhamnosyl} (1\rightarrow6) - \beta - D - \text{Glucose}]\)

Neohesperidose \([\alpha - L - \text{Rhamnosyl} (1\rightarrow2) - \beta - D - \text{Glucose}]\)
Flavonoids are widely distributed among the dicotyledonous angiosperms. Some classes are more widely distributed than others; while flavones and flavonols are universal, isoflavones and biflavonyls are found only in a few plant families. Although there are over a hundred flavonol aglycones known, only three are at all common; kaempferol (XXIIb), quercetin (XXb) and myricetin (XXXIVb). The other known flavonols are mostly structural variants of the common flavonols and are of limited occurrence. Flavonoids are present in plants as mixtures and it is very rare to find only a single flavonoid component in a plant tissue. There are often mixtures of flavonoid classes [Harborne, 1984].

Sophorose [β – D – Glucosyl (1→2) – β – D – Glucose]
(a) $R^1, R^2, R^3, R^4 = H$, Gossypetin

(b) $R^1 = \text{Glu, } R^2, R^3, R^4 = H$, Gossypetin 3 – O – glucoside

(c) $R^1 = \text{SO}_3\text{H, } R^2, R^3, R^4 = \text{Glc}, \text{Gossypetin 3 – sulphate 8 – O – glucoside}$

(d) $R^1, R^2, R^4 = H, R^2 = \text{Rha, Gossypetin 8 – O – rhamnoside}$

(e) $R^1, R^3, R^4 = H, R^2 = \text{Glc, Gossypetin 8 – O – glucuronide}$

(f) $R^1 = \text{Mal – Rut, } R^2 = \text{Me, } R^3, R^4 = H$, Gossypetin 8 – methoxy –3 – malonylrutinoside

(g) $R^1 = \text{Rut, } R^2, R^3 = \text{Me, } R^4 = H$, Gossypetin – 3’,8 – dimethyl ether 3 – O – rutinoside

(h) $R^1 = H, R^2, R^3, R^4 = \text{Me, Gossypetin – 3’, 4’, 8 – trimethylether.}$

The quantity of flavonoid formed varies in many plants from internal causes independent of external conditions. Flavonoids may originate at the point of vegetation but are more frequently formed in older tissues. Each organ and each tissue may carry flavones. External factors such as starvation and special effect of potassium and magnesium ions may cause variation only in extreme cases. The greatest variations occur at the transition periods of three phases of growth. In the older woody tissue the change generally occurs at the time of death. Flavonols may be present as a precursor of flavones and anthocyanins may be formed from the latter [Klein and Werner, 1925].
The flavonoid aglycones usually occur externally on the plant surface. Presently names of some twenty families are known which have accumulated flavonoid aglycones externally [Woolenweber and Jay, 1988]. Their lipophilic nature excludes their accumulation in the aqueous environment of the cell sap and this leads to external accumulation as a consequence of glandulotropic or epidermal
secretion [Hansel, 1962; Rimpler, 1965]. Flavonoid aglycones are hence encountered in oil cells or oil cavities, in and on glandular trichomes, in bud excretions, in thin epicuticular layers on leaves and in leaf wax and leaf resins. Glycosides are found in aqueous environment of cell sap. Dihydroquercetin (XXXVI) in the glycosidic form is a well – known heartwood constituent of many trees [Harborne, 1973].

In a recent report on flavonol glycoside production in callus cultures of Epimedium diphylllum it has been noted that effects of harmonal factors on cell growth and flavonol glycoside production indicated that 2, 4 – dichlorophenoxyacetic acid (XXXVII) was needed for the production of flavonol glycosides [Yamamoto et al., 1992].

![Chemical structure](image)

**XXXV**

(a) \( R = H, \) Isorhamnetin  
(b) \( R = \) Neohesp, Isorhamnetin \( 3-O - \) neohesperidoside  
(c) \( R = \) Glu, Isorhamnetin \( 3-O - \) glucoside  
(d) \( R = \) Gal, Isorhamnetin \( 3-O - \) galactoside  
(e) \( R = \) Rut, Isorhamnetin \( 3-O - \) rutinoside.
Structural features of flavonoids known to be of importance in function are (i) presence of a resonating system often in extended conjugation with a carbonyl chromophore which is responsible for colour and the pigments in plants (ii) presence of aromatic hydroxyls – depending on whether B – ring is monohydroxylated at C – 4’ or is of catechol type viz. carrying hydroxyls at C – 3’ and C – 4’ means that flavonoids are capable of alternately inhibiting or stimulating certain enzyme systems (3) molecular shape – their physiological activity can be accounted for by their similarity in structure to animal hormones [Harborne, 1973].

Flavonoids, because of the presence of many hydroxyl groups, are easily attached to enzyme surfaces and are very potent inhibitors of some enzyme systems. Presence of flavonoids in plant extracts often makes isolation of enzymes very difficult. The possibility that flavonoids act as growth regulators in plants by their effect on indole acetic acid – indole acetic acid oxidase enzyme system has been extensively explored [Galstonon, 1969].
The presence of flavonoids in spinach chloroplasts [Ottmeir and Heupel, 1971] and the isolation of kaempferol 3 – O – arabinoside (XXIc) from the chloroplasts of Impatiens balsamina [Weissenboeck et al., 1971] have suggested that the flavonoids may play a role in the photosynthesis of plants.

Some evidence has been presented for the flavonoid compounds having a function in the sexual process of the plant. The inability of two varieties of Forsythia to cross – pollinate is associated with the presence of rutin in the pollen of one and quercitrin (XXc) in the other [Kuhn and Low, 1949].

Flavonoid compounds act as substrates for redox processes relating to the function of phenolase [Szent – Gyorgyi and Vietorisz, 1931]. Flavonoids have been shown to be effective in preventing fungal attacks [Pridham, 1960].

The remarkable efficacy of highly hydroxylated flavones in acting as antioxidants for fats has been established [Lea and Swoboda, 1956].

Of all the natural pigments that can be used as dye-stuffs the flavones are by far the most widely distributed in nature. Luteolin (XXXVIIIa), the main colouring matter of the herbaceous plant weld Reseda luteola is the oldest European dye – stuff known. Dye – stuffs like weld, young and old fustic and quercitron bark are some of the flavone dye-stuffs which are still significant economically [McGraw Hill, 1966].

Most of the ultraviolet absorbance due to solar irradiation is attributable to flavonoids [Cadwell, 1971]. Flavonoids play subtle role as filters in plant growth.

Certain flavonoid glycosides like naringin (XXXIXa) and neohesperidin (XLa) are intensely bitter. The specific linkages in the sugars present are reported to be the determining factors for their bitterness [Kanniya, 1974].

The pharmacological importance of flavonoids came to light when it was found that crude preparation of vitamin C (XLI) derived from natural sources were far more active than the pure vitamin C alone in curing capillary lesions [Ruszyak and Szent, 1936].
(a) $R^1, R^2, R^3, R^4, R^5 = H$, Luteolin

(b) $R^1, R^3, R^4, R^5 = H, R^2 = \text{Glu}$, Luteolin 7 – O – glucoside

(c) $R^1, R^3, R^5 = H, R^2 = \text{Rut}, R^4 = \text{Me}$, Chrysoeriol 7 – O – rutinoside

(d) $R^1, R^3, R^5 = H, R^2 = \text{Glu}, \text{Api}, R^4 = \text{Me}$, Chrysoeriol 7 – O – glucoapioside

(e) $R^1, R^2, R^3, R^5 = H, R^3 = \text{Glu}$, Orientin

(f) $R^1 = \text{Glu}, R^2, R^3, R^4, R^5 = H$, Isoorientin

(g) $R^1, R^2, R^4 = H, R^3 = O \text{Glu}, R^5 = \text{Me}$, Hypolaetin 4' – methylether – 8 – O – glucoside

(h) $R^1, R^2, R^3 = H, R^3 = O \text{Glu}, R^4 = \text{SO}_3\text{H}$, Hypolaetin 8 – O – glucoside – 3’ – sulphate

(i) $R^1, R^3, R^4, R^5 = H, R^2 = \text{Glucur}$, Luteolin 7 – O – glucuronide

(j) $R^1, R^3, R^2 = H, R^2 = \text{Glu} – \text{Api}, R^4 = \text{Me}$, Chrysoeriol 7 – O – glucoapioside

Phenolic constituents usually improve the solubility characteristics of compounds and the phenols in general easily move across biological membranes [Parke, 1968]. So flavonoids have relatively weak influences on a broader range of biological phenomena since altered membrane characteristics appear to be a major means by which organisms control their biochemistry [Oaks and Bidwell, 1970]. Highly hydroxylated flavones act as diuretics [Koike, 1931; Fukuda, 1932].

Flavones especially eupatorin (XLII) isolated from Eupatorium spp. have been demonstrated to have cytotoxic and antineoplastic activities [Midge and Rao, 1975; Lee, 1977].
Quercitrin, kaempferol and naringenin (XXXIXb) isolated from Helichrysum angustifolium (syn. H. italicum) have been reported to increase bile secretion in experimental animals. Quercitrin also increased the detoxifying function of the liver and exhibited anti-inflammation activity [Prokopenko and Kiew, 1973].

(a) $R^1, R^3 = H$, $R^2 = \text{Neohesp, Naringin}$
(b) $R^1, R^2, R^3 = H$, Naringenin
(c) $R^1, R^2 = H$, $R^3 = \text{Xyl – Glu, Naringenin 4’ – O – xyloglucoside}$
(d) $R^1, R^2 = \text{Me}$, $R^3 = \text{Xyl – Ara, Flavanone 5, 7 – di – O – methylether 4’ – O – xyloarabinoside}$

(a) $R = \text{Neohesp, Neohesperidin}$
(b) $R = H$, Hesperetin
The flavonoids hesperetin (XLb) and rutin in peppermint leaves have choleretic activity in dogs [Pasechnik et al., 1967]. The flavonoids present in Passiflora incarnate reportedly have tranquilizing effects [Lutomski, 1975]. Isoflavones like genistein (XLIIIa) and daidzein (XLIIIb) have been found to be oestrogenic. The molecular shape of genistein is similar to that of oestradiol (XLIV) [Biggers, 1960; Harborne, 1967]. The role of flavonoids in such clinical conditions such as hypertension, rheumatism, arthritis and pregnancy as also their beneficial effects in frost – bite and cold injury have been studied [Subramanian and Nair, 1963].

Cassia alata leaf extract which is known for its polyphenolic constituents has been recently studied for its wound – healing activity and mast cell stabilization [Palanichamy et al., 1991; Palanichamy et al., 1991a].

Certain flavonoids like quercetin have been found to decrease capillary permeability induced by histamine (XLV) [Paris and Vaivel, 1949]. By prior administration of flavonoids the increase in permeability produced by heat, X – ray and UV radiations could be reduced [Bohr et al., 1949 and Parmar and Gosh, 1977]. Flavonoids have also been found to increase capillary resistance [Highly and Middleton, 1942]. The efficacy of the flavonoids in capillary fragility has been claimed [Biggers, 1960]. Small quantities of flavonoids act as cardiac stimulants but in larger doses systolic stoppage results [Fukuda, 1932]. Some flavonoids appear to strengthen weak capillary blood vessels and hence are of interest in the search for means of counteracting the evils of exposure to radioactivity [Clark and Geissman, 1949].
Eupatorin

(a) $R_1, R_4 = OH, R_2, R_3 = H$, Genistein
(b) $R_1, R_2, R_3 = H, R_4 = OH$, Daidzein
(c) $R_1, R_3 = H, R_4 = OH, R_2 = Glu$, Daidzein 7 – O – glucoside
(d) $R_1, R_4 = H, R_2 = Me, R_3 = OH$, 3’ – hydroxyl – 7 – methoxyisoflavone
(e) $R_1 = H, R_3, R_4 = OH, R_2 = Rha – Glu$, 3’ – hydroxydaidzein 7 – O – rhamnoglucoside
(f) $R_1 = H, R_3, R_4 = OH, R_2 = Xyl – Glu$, 3’ – hydroxydaidzein 7 – O – xyloglucoside
Several flavonoids are moderately effective against laboratory cultures of malignant cells. Quercetin pentamethyl ether (XXd) and rutin have been shown to be strongly and moderately effective in inhibiting respectively of benzopyrene induced pulmonary adenoma in mice. In these cases the flavonoids may induce benzopyrene hydroxylase which may detoxify carcinogen. Antiulcer [Vogin and Rossi, 1961] and antitumor [Kupchan et al., 1969] activities of flavonoids have been observed.

Flavonoids and their related compounds synthesized in plants possessing antiviral, antifungal and bacteriostatic action are absorbed into the body and they attack reversibly the blood cells. In addition to any potential action against pathogens, certain of these compounds potentiate enzymes which detoxify carcinogenic hydrocarbons, exhibit anti – inflammatory activity, exert anti – adhesive action on blood cells and show anti-thrombogenic activity [Robbins, 1975].

Studies on toxicity to fish of some chalcones, flavonols and flavones have been recently carried out and it has been reported that a concentration of 1% (w/v) as dioxane solution is harmless to the fish over a period of 24 hrs and that halogenoflavonols are more toxic than their corresponding chalcones [Subbanwad et al., 1991].
Earlier reports on the identification of known flavonoids in new plant sources as well as those on novel flavonoids have been summarized [Harborne, 1988]. General plant survey for anthocyanins [Lawrance et al., 1939] and flavonols [Bate–Smith, 1962] has also been done.

General complexity in pattern increases with evolutionary advancement whereas morphologically primitive groups such as mosses, horsetails and ferns have simple flavonoids and the more highly specialized angiosperms have much more complex flavonoid constituents. So, monocotyledons and dicotyledons show significant differences in their flavonoid patterns [Harborne, 1973].

Of the many and varied classes of secondary constituents, flavonoids have probably attracted the most attention as taxonomic markers in plant systematics [Erdtman and Swain, 1963].

\[ \text{XLVI} \]

(a) \( R^1, \ R^2 = H, \) Cyanidin  
(b) \( R^1 = \text{Rut}, \ R^2 = H, \) Cyanidin 3 – O – rutinoside  
(c) \( R^1 = \text{Glu}, \ R^2 = H, \) Cyanidin 3 – O – glucoside  
(d) \( R^1 = \text{Caffeylrha – glu}, \ R^2 = H, \) Cyanidin 3 – O – Caffeyl – rhamnosylglucoside  
(e) \( R^1 = \text{Caffeylrha – glu-glu}, \ R^2 = H, \) Cyanidin 3 – O – Caffeyl – rhamnosyldiglucoside  
(f) \( R^1 = \text{Caffeylglu}, \ R^2 = H, \) Cyanidin 3 – O – Caffeylglucoside  
(g) \( R^1 = \text{Caffeylrut}, \ R^2 = H, \) Cyanidin 3 – O – Caffeylrutinoside  
(h) \( R^1 = \text{Gal}, \ R^2 = H, \) Cyanidin 3 – O – galactoside  
(i) \( R^1, \ R^2 = \text{Glu}, \) Cyanidin 3, 5 – O – diglucoside  

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(a) $R^1, R^2 = H$, Delphinidin
(b) $R^1 = \text{Rha} - \text{Glu}, R^2 = \text{Xyl}$, delphinidin 3 – O – rhamnosyl glucoside – 7 – O – xyloside

(a) $R^1, R^2 = H$, Pelargonidin
(b) $R^1 = \text{P – Coumarylglu}, R^2 = H$, Pelargonidin 3 – O – P – Coumarylglucoside
(c) $R^1 = \text{Rha} – \text{Glu} – \text{Glu}, R^2 = H$, Pelargonidin 3 – O – rhamnosyldiglucoside
(d) $R^1, R^2 = \text{Glu}$, Pelargonidin 3, 5 – O – diglucoside.

The change over from woody to herbaceous habit in the angiosperms has caused a corresponding change in flavonoid pattern chiefly in the leaves. The woody pattern is characterized by high concentration of flavonol, presence of myricetin and leucoanthocyanidins and frequent occurrence of cyanidin (XLVIIa). By contrast, the herbaceous pattern is shown by infrequency of flavonol, almost complete absence of
myricetin and leucoanthocyanidins, presence of flavones and frequent occurrences of delphinidin (XLVIIa) and pelargonidin (XLVIIIa) as flower pigments [Harborne, 1973].

The Asterales (Asteraceae) give rise to phenomenal array of flavonoids. It comprises of 1,100 genera and 25,000 species [Heywood, 1979]. The flavonols quercetin and kaempferol are common especially, the polymethylated types including compounds with 6 – and 8 – hydroxyl / methoxyl substitution. Similar patterns are observed among flavones. Carbon – carbon linked glycosides are rare. Asteraceae is the only family to experiment, extensively with anthochlors. In this respect Asteraceae have concentrated 'exploitation' of flavonoids (many with biological activity), sesquiterpene lactones (also with biological activity) and polyacetylenes and in a few cases alkaloids. Myricetin and several methylated derivatives have recently been isolated in Haplopappus [Avanoglu et al., 1981].

Flavone glycosides isolated from the flowers of Centaurea aspera [Ferreres and Tomas, 1988]. C. cyanus [Tamura et al., 1983] and Chrysanthamum indicum [Chatterjee et al., 1981] are apigenin 7 – (6" – ethylglucuronide) (XXIf), apigenin 4′ – (6" – malonylglucoside) – 7 – glucuronide (XXIg) and acacetin 7 – galactoside (XXIh) respectively. 3′, 4′, 5, 7 – tetrahydroxy – 6 – C – prenylflavanone (XLIXa) has been isolated from Wyethia angustifolia [Mc Cormick et al., 1986] W. arizonica [Bohlmann et al., 1984], W. glabra [Mc Cormick et al., 1985] and W. helenioides [Bohlmann et al., 1981]. The corresponding 8 – C – analogue (XLIXb) has been reported from W. angustifolia.

Dihydroflavonols like padmatin (La) and 3 – acetyl padmatin (Lb) have been identified in Artemisia glutinosa [Gonzalez et al., 1983] and Inula viscosse [Grande et al., 1985] respectively. Coreopsis (LI) has been found in Coreopsis [Crawford and Smith, 1985] Helianthus heterophyllus [Schilling, 1983] and Viguiera dentate [Reiseberg and Schilling, 1985]. From Bidens torta [Mc Cormick et al., 1984] 2′, 3, 3′ – trihydroxy 4 – methoxychalcone – 4′ – glucoside (LIIa), 2′, 3, 3′ – trihydroxy 4 – methoxychalcone – 4′ – glucose – X" – acetate (LIIb) and 2′, 3′ – dihydroxy 3, 4 – dimethoxychalcone – 4 – glucoside (LIIc) have been recorded.
(a) $R^1 = \text{Prenyl, } R^2 = \text{H, } 3', 4', 5, 7 - \text{tetrahydroxy – 6 – prenylflavanone}$

(b) $R^1 = \text{H, } R^2 = \text{Prenyl, } 3', 4', 5, 7 - \text{tetrahydroxy – 8 – prenylflavanone}$

(a) $R^1, R^3, R^4 = \text{H, } R^2 = \text{Me, Padmatin}$

(b) $R^1 = \text{OOCCH}_3, R^2 = \text{Me, } R^1, R^4 = \text{H, } 3 - \text{Acetylpadmatin}$

Anthoxanthins (flavonols, flavones, glycoflavones etc.) are commonly produced in leaves and flowers of Caryophyllales coexisting with the betallins. An early survey by Bate Smith (1962) has established the presence of flavonols,
absence of ellagic acid and limited occurrence of myricetin in the Caryophyllales [Burrett et al., 1982].

\[
\begin{align*}
&\text{LII} \\
&\text{(a) } R^3 = \text{Glu, } R^1, R^3 = \text{H, } 2', 3, 3' - \text{trihydroxy 4 - methoxychalcone} \quad 4' - \text{glucoside} \\
&\text{(b) } R^2 = \text{Glu} - X'' - \text{acetate, } R^1, R^3 = \text{H, } 2', 3, 3' - \text{trihydroxy 4 - methoxychalcone} \quad 4' - \text{glucose} \quad X'' - \text{acetate} \\
&\text{(c) } R^2 = \text{Glu, } R^3 = \text{Me, } R^1 = \text{H, } 2', 3' - \text{dihydroxy 3, 4 - dimethoxy chalcone} \quad 4' - \text{glucoside} \\

\text{Four flavanones Isosakuranetin (IIIc), Persicogenin (IIIId), 5, 6, 7, 4' - tetramethoxy flavanone (IIIe), 4' - hydroxyl - 5, 6, 7 - trimethoxy flavanone (IIIf), 2' - Hydroxy - 4, 4', 5', 6' - tetramethoxychalcone, 4, 2' - dihydroxy - 4', 5', 6' - trimethoxychalcone and two flavones Acacetin (5, 7 - dihydroxy - 4' - methoxyflavone) and Luteolin (5, 7, 3', 4' - tetrahydroxy flavones) have been reported from the flowers of Chromdaena odorata and analysed for their antibacterial activity and cytotoxicity [Apichart Suksamrarn et al., 2004]. Jaceosidin (IVc) and Cirsimicolic (IVd) have been isolated from the aerial part of Centaurea pullata [Medjroubi et al., 2005]. From the aerial part of Centaurea tougourensis, 5, 7, 4' - trihydroxy - 6, 3' - dimethoxy flavone (Jaceosidin), 5, 7 - dihydroxy - 6, 3', 4' - trimethoxyflavone (IVe), 5 - hydroxyl - 6, 7, 3', 4' - tetramethoxyflavone (IVf), 5, 7, 3', 4' - tetrahydroxy - 6 - methoxyflavone (IVg), 5, 7, 4' - trihydroxyflavone (apigenin), 3, 5, 7, 4' - tetrahydroxyflavone (Kaempferol) have been isolated and identified [Nacer et al., 2006].}
\end{align*}
\]
The flavonoids namely 5, 7, 4’ – trihydroxy – 8 – methoxyflavone (isooscutellarein – 8 – Me – ether), 5, 3’, 4’ – trihydroxy – 7 – methoxyflavone (luteolin 7 – Me – ether), 5, 3’, 4’ – trihydroxy – 7 – O – glucosyl flavone (luteolin 7 – glucoside), Apigenin 8 – C – glucoside (vitexin) and 5, 7, 4’ – trihydroxy – 3’ – methoxy – 3 – O – glucosyl flavone (cacticin) have been reported from the aerial part of Chrysanthemum fuscatum. All these compounds were isolated for the first time from C.fuscatum. The n-butanol extract of C.fuscatum was evaluated for hepatoprotective activity in rats with acute hepatitis induced by isoniazid and rifampicin. The extract in an oral dose (200 mg/kg body weight) exhibited a significant protective effect by lowering lipid peroxidation and by enhancing the glutathione system [Ameddah et al., 2007].

Isorhamnetin – 3 – O – rutinoside (XXXVe) (narcissin) and quercetin – 3 – O – β – D – glucopyranoside (Isoquerцитrin) have been reported from the flowers of Calendula officinalis. Compounds were isolated for the first time in the Russian Federatim from cultivated medicinal Calendula flowers [Kurkin and Sharova, 2007]. From the aerial parts of Centaurea furfuracea, Isokaempferide 7 – O – methyl glucuronide and Isokaempferide 7 – O – Glucuronide have been identified and antiplasmodial and cytotoxic activities have been evaluated [Akkal et al., 2007]. A new 2 – phenoxychromone glycoside has been isolated from the n-butanol extract of Artemisia rupestris and reported as 6 – demethoxy – 4’ – O – methylcapillarisin – 7 – O – β – glucoside [Zhao et al., 2009].

5, 6, 7 – trihydroxy – 4’ – methoxyflavonone (IIIg) has been reported from the leaves of Baccharis retusa DC. It is active against Leishmania sp and Trypanosoma cruzi [Grecco et al.,2010] 5, 4’ – dihydroxy – 3’, 6, 7 – trimethoxyflavone (Cirsineole), 5, 7, 4’ – trihydroxy – 6, 3’ – dimethoxy flavone (Jaceosidin), 5 – hydroxy – 6, 7, 3’, 4’ – tetramethoxyflavone (3’ – O – methyl eupatorin), 5, 7, 3’, 4’ – tetrahydroxy – 6 – methoxyflavone (nepetin) and 5, 7-dihydroxy – 6, 3’, 4’ – trimethoxyflavone (eupatilin) have been isolated and identified from the aerial parts of Centaurea sulphurea [Kabouche et al., 2011]. Along with Sesquiterpene lactones, flavonoids, Centaureridin and Penduletin have been reported from the aerial parts of Anthemis rutheriana [Vujisic et al., 2011].
From the aerial parts of the flowering plant Centaurea omphalodes, Chrysin, Cirsimaritin, Salvigenin, Chrysoeriol and Chrysin 8 – C – Glc are isolated. All the compounds are reported for the first time from the species [Khalfallah et al., 2012]. Luteolin (XXXVIIIa) has been isolated from the leaves of Cichorium endivia L. subsp. Divaricatum. [Nadia M. El-Shafey et al., 2012]. From the aerial parts of Cotula cinerea seventeen flavonoids have been isolated and identified as chrysospenol – D, chrysosplenatin, oxyayanin – B, axillarin, 3 – methylquercetin, pedalenin, isokaempferid, apigenin, luteolin, 6 – hydroxyluteolin (XXXVIIIk), 3 – glucosylisorhamnetin (XXXVc), methyl – 7 – glucosylquercetin, 7 – O – α – D – glucosylapigenin, 7 – O – β – D – glucosylluteolin, 7 – O – β – D – glucosylquercetin, 7 – O – α – D – glucosylaxillarin, 7 – O – β – D – diglucosylluteolin. Among these seventeen flavonoids twelve compounds were not previously reported for the genus Cotula [Dendougui et al., 2012]. A flavonoid C – glycoside ie, namely 5 – hydroxy – 7 – methoxy – 6 – C – glycosyl – flavone has been isolated from the aerial part of S. indicus [Mishra et al., 2007]. A novel isoflavone glycoside, 5, 4′ – dimethoxy – 3′ – prenylbiochanin 7 – O – β – d galactoside has been isolated from the leaves [Yadav and Kumar, 1999]. A flavone glycoside, 7 – hydroxy – 3′, 4′, 5, 6 – tetramethoxy flavone – 7 – O – β – d – (1 – 4) diglucoside has been isolated from the stem of S. indicus [Yadav and Kumar, 1998].

The Rutaceae, as a family is extremely versatile in its synthetic capacity and is recorded to produce a wide range of unusual, highly substituted flavonoid constituents. Flavonoids of this family are characterized by a high degree of methylation, by extra substitution in the 6 –, 8 – or 2′ – position, by the presence of methylenedioxy groups and by attachment of isoprenoid sidechains. Citrus spp. apparently contains several glycocflavones; vitexin and xylosylvitexin (VIIIId) in which the xylene is attached β (1→2) to the C – 8 of the apigenin molecule have also been reported.

A novel flavonol viz. phellamuretin (LIII) has been obtained from Phellodebdron japonicum [Grimshaw and Lamer, 1972] whereas the leaves of Murraya paniculata have been found to contain 3, 3′, 5, 5′, 6, 7 – hexa methoxy flavone (LIVa). From the fruits of Murraya omphalocarpa 3, 3′, 4′, 5, 5′, 6, 7 – heptamethoxyflavone (LIVb) has been identified [Wu et al., 1980].
Aerial parts of Halophyllum suaveolens and H.buxbaumi have been found to contain gossypetin 8 – methylether – 3 – malonylrutinoside (XXXIf), quercetin 3 – glucoside (XXe) and isorhamnetin 3 – glucoside (XXXVc). In addition, gossypetin 3’, 8 – dimethylether – 3 – rutinoside (XXXIIg), isorhamnetin 3 – O – rutinoside (XXXVe) and apigenin 7 – glucoside have been characterized from H. suaveolens while quercetin, isorhamnetin (XXXVa) and gossypetin 3’, 4’, 8 – trimethylether (XXXIIh) have been found in H. bauxbaumi [Ulubelen, 1986].

(a) R = H, 3, 3’, 5, 5’, 6, 7 – Hexamethoxy flavone

(b) R = OMe, 3, 3’, 4’, 5, 5’, 6, 7 – Heptamethoxy flavone

From Citrus plants, two flavonoids, 3, 5, 6, 7, 8, 3’, 4’ – heptamethoxy flavone and natsudaidain have been isolated and studied as cardiotonic flavonoids [Itoigawa et al., 1994]. A flavanone glycoside, Brutieridin (LV) has been discovered in bergamot orange juice and exhibits statin-like properties. In the citrus fruits, Hesperidin has been found abundantly. It has also been reported in the fruit of Citrus qurantium L (Bitter orange), Citrus sinensis, Zanthoxylum qilletii and leaves
of Aqathosm aserratifolia. Isosakuranetin, a flavanone, has been found in the fruit of Citrus sinensis, in the fruit of Citrus x. paradisi and in Monarda didyma. From the roots of Citrus sinensis, 5, 8 – dihydroxy – 6, 7, 4′ – trimethoxy flavone has been identified. The 7 – O – neohesperidoside of isosakuranetin has been known as Poncirin (LVI) which has been isolated from Poncitrus trifoliate [Intekhab and Aslam, 2009].

From bergamot orange juice, a flavanone glycoside melitidin has been reported. A flavanone glycoside naringin, a major flavonoid in grapefruit has been responsible for the bitter taste of the fruit juice. Natsudaidain, an O-methylated flavonol, has been isolated from citrus plants. The name of the isolate comes from Citrus natsudaidai (Natsumikan lit. “summer tangerine”), a fruit of Japan. An O –
methylated flavone nobiletin isolated from citrus peels like in tangerine has been found to have anti – inflammatory, anti – tumor invasion, proliferation and metastasis in vitro and in vivo. It has also been found potentially to inhibit cartilage degradation. Tangeritin, an O – poly methoxylated flavone which has been found in tangerine and other citrus peels, acted as plants defensive mechanism against disease causing pathogens. Zapotin, a flavone isolated from casimiro aedulis has been found to possess potential anti – carcinogenic effects against isolated colon cancer cell.

The Convolvulaceae, comprising of 50 genera and 1000 species has been recorded to contain many common flavonoids. It is one of the 24 plant families from which zwitter ionic anthocyanins have been recorded [Harborne, 1986].

LVII Heavenly blue anthocyanin

LVIII 3',4',5,7-Tetrahydroxy flavone

Petals of Ipomoea purpurea [Katoka, 1939; Goto, 1984; Goto et al., 1881] have been recorded to contain pelargonidin 3, 5 – O – diglucoside and heavenly blue anthocyanin (LVII). A new flavone viz. 3', 4', 5, 7 – tetrahydroxy flavone (LVIII) has been reported [Gupta et al., 1984] from the whole plant of Evolvulus nummularius. Kaempferol 3 – O – galactoside (XXIk), kaempferol 3 – O –
rutinoside (XXIf), kaempferol 3 – O – glucoside (XXIe) and quercetin 3 – O – rutinoside have been isolated from the leaves of Calystegia hederaceae, leaves of C. japonica [Hukuti, 1939], stem of Cuscuta reflexa [Subramanian and Ramanathan, 1963] and tubers of Ipomoea spp [Harborne, 1967]. 3 – O – Glucoarabinosides of quercetin (XXm) and fisetin (LIX) [Valsakumari, 1990] and isorhamnetin 3 – O – neohesperidoside (XXXVb) [Dhandapani and Nagarajan, 1989] have also been reported from the flowers of C. reflexa. Kaempferol and its 3 – O – glucoside (astragallin) have been isolated from the flowers of Ipomoea aquatica [Sukumar, 1989] and I. carnea [Sukumar and Sulochana, 1987]. Kaempferol 3, 7 – O – di – O – glucoside (XXIII) has also been reported from the petals of I. carnea [Nambi, 1989].


species. Quercetin 3 – O – galactoside 3 – O – glucuronide and Kaempferol 3 – O – galactoside 7 – O – glucoside isolated and identified from leaves and Quercetin 3, 7 – O – digalactoside from stems of C. japonica [Ahn et al., 2012]. Four flavonoids, diosmetin, luteolin and diosmetin – 7 – O – glucoside have been isolated from the aerial parts of Merremia tridentate Hallier. The plant has been reported to possess antidiabetic, anti – intflammatory, anti – arthritis, wound healing, analgesic and antimicrobial activities [Neyanila et al., 2013].

Cucurbitaceae comprises of about 100 genera and over 750 species. There is a tremendous genetic diversity within the family and the range of adaptation for cucurbitaceae species includes tropical and subtropical regions, arid desert and temperate areas. They are recognized as source of secondary metabolites and hence possess pharmaceutical values as anti-HIV, antiulcer, anti-inflammatory, antileukemia, antimicrobial, antidiabetic and antitumour activities [Amira et al., 2003; Mazza et al., 2001].

The O–Glycosylisovitexins have been reported from Cucumis melo [Monties, 1971]. From Bryonia dioica, 7–O–glucosyl 8–C–glucosyl apigenin (Alliaroside) has been isolated [Paris et al., 1966; Leiba et al., 1968]. 7–O–glucosyl–6–C glycoside apigenin, 6–C–glucoside apigenin, 6–C–glycoside luteolin and 7, 4’– O-diglucosyl –6–C– glucoside [Rakesh et al., 2010] and saponarin 4’ – O – glucoside have been obtained from the Lagenaria siceraria. Kaempferol and quercetin–3–O–rutinoside have also been isolated from the pollen of Lagenaria siceraria [Gangwal et al., 2010].

From Trichosanthes kirilowiivar, 7–3’– and 4’– glycosides of luteolin, 7–glucoside and 6, 8– di–C–glucoside of apigenin have been isolated. Kaempferol 3, 7–di–rhamnoside and 3–glucoside–7–rhamnoside obtained from T. cucumeroides and Kaempferol 3–galactoside and 3– sophoroside have been detected from T. anguina. Quercetin 3 – O – rutinoside has been detected from T.multiloba and T. rostrata. From T. bracteata, luteolin 3’–O –glucoside and kaempferol 3–O–rutinoside have been isolated. Luteolin 7, 3’– and 4’–O–glucosides and apigenin 7–O–glucoside have been afforded from T. kirilowii [Yoshizaki et al., 1987].
Isovitexin 2’- O-glucoside, isoritexin, isoorientin, 4’-O-diglucosides of isovitexin and swertiajaponin has been isolated from the leaves of Cucumber ativus. From the flowers of the above species, kaempferol 3-O-rhamnoside and 3-O-glycosides of kaempferol, quercetin and isoramnetin have been reported [Baranowska et al., 2001]. A series of kaempferol and quercetin glycosides occurred as 3 – O – glycosides, 3 – 7 – di – O – glucosides, 3 – O – glucosides – 7 – O – rhamnosides, 3 – O – diglucosides and as 3, 7 – substituted triglycosides from Marah oreganus [Nicholls and Bohm, 1982]. Eight flavonoids, including three C – glucosyl and five O – glucosyl flavones have been reported from Sechiisimule (Jacq) Swartz [Tiziana Siciliano et al., 2004]. From the extract of Cayaponia tayuya roots, Vicenin – 2 spinosin, isovitexin, swertisin and isoswertisin have been reported [Aquila et al., 2009]. From the aerial parts of Sicyos angulatus, seven flavonol glycosides were isolated and identified as 3, 7 – O – glycosides of quercetin and kaempferol [Na et al., 2013].

Papaveraceae is an economically important family of about 44 genera and an approximately 770 species of flowering plants in the order Ranunculales. The family is cosmopolitan, occurring in temperate and subtropical climates, but almost unknown in the tropics. Most are herbaceous plants, but a few are shrubs and small trees.

Cyanidin 3 – rhamnosylglucose, 3 – rhamnosyl – glycoside – 7 – kyloside and Cyandin 3, 5 – diglucoside have been isolated from the flower and leaf of Dicentra spp. / hybrids [Fahselt, 1968]. From the petal of Meconopsis paniculata, 8 – Hydroxy kaempferol (Herbacetin) 3 – glucoside has been isolated [Harborne, 1969 a].

Quercetin 3’ – methylether (Isorhamnetin) 3 – O – glucoside has been observed from the flower of Argemone Mexicana [Rahman and Ilyas, 1961]. Krishnamurthi et al.,(1965) isolated Quercetin 3’ – methylether 3, 7 – diglucoside. Cyanidin – maloxylsambubioside – 7 – glycoside, Kaempferol 3 – gentiobioside and kaempferol 3 – xylosylgentiobioside are isolated from Meconopsis betonicifolia [Takeda et al., 1996]. Seven flavonoids, quercetin, dihydroquercetin, luteolin, chrysoeriol, apigenin, huazhongilexone and hydnoacarpin are isolated and reported from Meconopsis quintuplinervia regel [Xiaoya Shang et al., 2002].
Isorhamnetin 3 – O – arabinosyl (1→6) – glucoside is identified from the leaves of Papaver orientate. Kaempferol has been reported in Meconopsis integrifolia flowers and in Fumaria parviflora, kaempferol 3 – O – gentiobioside, 3 – O – rutinoside and 3 – O – glucoside – 7 – O – galactoside are identified. The three C – glycosides, Vicenin – 2, (6, 8 – di – C – glucosylapigenin), Schaftoside (6 – C – glucosyl – 8 – C – arabinosyl apigenin) and isoschaftoside (6, 8 – di – C – glucosyl apigenin) in Roemaria hybrid are reported. Herbacetin and gossypetin 3 – O – glucuronide – 8 – O – glucosides are also reported as new glycosides in R.hybrida. In the petals of Papaver nudicaule Gossypetin – 7 – O – glucoside has been identified [Saleh et al., 1988].
AIM AND SCOPE

The present study is an attempt to screen various phytochemical constituents like alkaloids, flavonoids, flavones, flavanones, flavonols, isoflavones, glycosides, tannins, phenolics, aminoacids, steroids, proteins, carbohydrates, saponins, terpenoids, fats/oils, anthocyanins and leucoanthocyanins and estimate total phenol, flavonoid, flavonol and tannin from the following medicinal plants.

a. Sphaeranthus indicus Linn.
b. Zanthoxylum tetraspermum Wight.
c. Cuscuta reflexa Roxb.
d. Citrus colycynthis (Linn.) Schrad.
d. Argemone mexicana Linn.

The study also aims at isolation of flavonoid pigments from the above mentioned species and determination of their detailed structures with modern techniques like UV, FTIR, \(^1\)H and \(^{13}\)C- NMR, Mass and chromatography.

Any scientific discovery becomes relevant only if it is augmented with applications for human emancipation. So, the study also projects at exploration of antioxidant and antimicrobial activities of the plant isolates and crude extracts.