CHAPTER I
INTRODUCTION

1.1 CANCER
1.1.1 Cancer

Cancer is a disease that is caused when the set of normal cells in our body changes its structure and leads to uncontrolled growth of cells which forms a swelling called tumour. This is common in all types of cancer except in the case of leukaemia. If it is not treated on time, the tumour can spread to the surrounding healthy tissues and other body parts through the blood stream, lymphatic system and can reach up to the digestive system, circulatory system and the nervous system. The malignant neoplastic disease can start in any part of the body like pancreas, colon, lungs, breast, oesophagus, stomach, or even in the blood stream. There are numerous types of cancers that may look alike. But from the way they develop to how they spread, every cancer is unique in its own way [1].

Every unit in our body has its unique function to perform. The human body has a rhythm and order of its own in cell division and development. Cancer occurs in the body when this rhythm of cell division gets affected where the cells loose the mastery over themselves. The result of which is uncontrolled manufacturing and development of new cells. The abnormal cells grow fast forming a cluster of themselves called tumour. Though all tumours generally increase in size, some tumours grow very fast where as others grow slowly. There are more than 200 types of tumours which are differentiated from one another in accordance with the types of affected cells [2].

Malignant neoplastic cancer where the abnormal cells mature rapidly has become so common nowadays (with an estimated average of one event of cancer among every four people in the United States) that a lot of families have at least a few affected persons. Approximately up to 15% of all cancers are genetic, which means that the cancer tend to occur among members of the same family. Varied cases of cancer occur by chance, or in accordance with common family habits like cigarette
smoking, tobacco consumption etc. The tumour that has spread from its place of origin to other parts of the body, resulting in the destruction of the healthy tissues all over the body is called metastasis. This stage is fatal where many patients are succumbed to death. In 2016, cancer has killed approximately 14.5 million people in the world. 8.2 million people lost their lives to cancer [3].

1.1.2 Types of Cancer

There are numerous types of cancers.

- Carcinoma, the most common type of cancer affects lung, colon, breast and ovaries.
- Sarcoma is common in bone, cartilage, fat and muscle.
- Lymphoma occurs in the lymph nodes of the body’s immune system. They comprise of the Hodgkin’s and Non-Hodgkin’s Lymphomas.
- Leukaemia is initialized in the bone marrow and later spreads in large numbers into the bloodstream [4].

1.1.3 Cancer Development

The intricate procedure through which the normal cells develop into cancer cells is called translation. In the first step called initiation, the genetic material in the DNA and the chromosomes gets altered, turning the cells malignant. This change may occur instantly or initiated by a cancer causing agent (carcinogen). The second step in the growth of cancer cells is called forwarding where the cell that has undergone initiation becomes cancerous. The factors that contribute to this process are called boosters or promoters, which can be either neighbouring cells or drugs. Unlike carcinogens, promoters do not persuade cancer by themselves. However, promoters allow a cell that has undertaken initiation to become cancerous. Promotion has no hand on non-initiated cells. Henceforth, installation and promotion are needed to cause cancer [5].

1.1.4 Pathology of Cancer

Tumours are classified as benign and malignant considering their mode of behaviour inside the human body. Benign tumours are more common in occurrence and less harmful except in the enclosed locations like the skull. As benign tumours can turn to be malignant they are usually removed as soon as they are diagnosed [6].
Malignant tumours are characterized by their fast growth. Their malignancy is very dangerous to the extent that every part of the patient is under threat. They have the capacity to attack surrounding cells and tissues as well as distant organs. One of the primary evidences of primary malignancy is the capacity of the tumour cells to separate itself from the primary tumour and setup a metastasis or a secondary tumour [7].

The survival rate of the cancer patients in the metastasis stage is very less and those who survive suffer from this side effects. The treatment cost of the modern medicine too is very high. These factors have served as a catalyst for the researchers all over the world to look out for better products which stands more in communion with nature and less in conflict with the human system. The possibility of using natural products as anticancer agents was recognized in the 1950’s by the US National Cancer Institute (NCI) and has done significant contributions to the discovery of new naturally occurring anticancer agents [8].

1.1.5 Causes of Cancer

The causes of cancers have been attributed to changes in the environment, lifestyle and genetic factors and no single cause. No single cause can be pointed out for the development of cancer. Major factors that causes death due to cancer are tobacco, diet and obesity (30-35%), infections (15-20%), radiation (both ionizing and non-ionizing up to 10%) [9].

1.1.5.1 Carcinogens

A carcinogen is any substance that sabotages a cell by altering its behaviour though mutation in the gene.

1.1.5.2 Age

The increase in the lifespan of the individual is directly proportional to the chance of getting exposed to carcinogens, which in turn causes genetic mutation in their cells [10].
1.1.5.3 Genetics

In many cases, people are born with a high risk for a specific cancer (genetic predisposition). This does not assure a cancer development, but a genetic predisposition makes the disease more probable.

1.1.5.4 Immune System

People with poor immune system are more at risk of imbibing malignant neoplastic cancer. This places people who have got organ transplants and under drugs to squash their immune system to prevent organ rejection, HIV positive individuals, and other medical conditions which suppress their immune system under high risk category of acquiring cancer.

1.1.5.5 Bodyweight, Diet and Physical Activity

Cancer researchers have found out that maintaining a healthy body mass index, following a healthy diet and doing regular exercises could avert at least one in three deaths due to cancer. Red and processed meat forms the stable diet of many with less importance given to fresh fruits and green veggies. This unbalanced diet increases the possibility of malignant neoplastic cancer.

1.1.5.6 Overweight or Obesity

Obese is being 25% overweight. Overweight or obese people are highly susceptible to bowel and pancreatic cancer, mainly due to a leaning towards higher insulin levels. Obesity can lead to risk of cancer in food pipe or Oesophagus (oesophageal cancer), kidney and gall bladder cancer, as well as breast or cervical cancer in females [11].

1.1.5.7 Alcohol

It is now proven that alcoholic drinks can elevate the risk of a number of cancers, including pharyngeal, laryngeal, oesophageal and liver cancers. Even the consumption of alcohol in mild dosages will increase the probability of the development of malignant neoplastic cancer.

1.1.5.8 Tobacco

Tobacco smoke has at least 80 varying carcinogenic agents. When the tobacco smoke is inhaled, the carcinogenic chemical agents enter the lungs, pass through the blood
and are carried throughout the human body. Hence, smoking or chewing of tobacco not only leads to lung and mouth cancers, but also is a gateway to numerous other cancers. Generally, a person starts smoking at a younger age which continues for a number of years making the person susceptible to all types of cancers.

1.1.5.9 Ionising Radiation

Artificial radiations can also cause cancer and are a threat for people working in conditions where they are exposed to radiations. Even the continuous exposure to the ultraviolet radiations from the sun, can lead to melanoma and skin malignancies. Fair complexion people and those with lots of moles who has already encountered melanoma or non-melanoma skin cancer, are the most vulnerable.

1.1.5.10 Workplace Hazards

Some people are more susceptible to cancer because of their profession. For example, workers in the chemical dye industry are more prone to bladder cancer. Asbestos is a known work place cancer causing agent - specifically a cancer called mesothelioma, which affects the pleura (covering) of the lungs.

1.1.5.11 Infection

There are certain types of cancers which are caused due to viral infection. This does not mean that all viral infections lead to cancer and are contagious. Rather it is caused due to the modifications in the cellular bodies initiated by the virus.

1.1.5.12 Signs and Symptoms

The symptoms of different types of cancer varies from one another and to a large extent depends on the location of the disease. A few primary signs and symptoms are as follows[12].

*Lumps*

Any lump in the human body can turn cancerous in due course of time. In most cases cancerous lumps increase in their size with time and are mostly painless. Some cancers can be felt through the skins.

*Coughing and Breathlessness*

Persistent coughing and breathlessness can be the indications of lung cancer.
Changes in the Bowel Habit

The symptoms of intestine cancer, in most cases will be the development of complications in the bowel movement like constipation, diarrhoea and blood in the commodes.

Bleeding

Unusual bleeding is an important indication of the presence of malignant neoplastic cancer.

- Haemorrhage from the anal passage may be a symptom of intestine cancer
- Haemorrhage from the cervix may be an indication of cervical cancer
- Haemorrhage in the urine may be a sign of kidney or bladder cancer

Unexplained Weight Loss

Huge unexpected weight loss over a short span of time (1-2 months) can be a major indication of cancer.

Fatigue

Fatigue is commonly experienced by the cancer patients along with other symptoms.

1.1.6 Tests for Cancer

1.1.6.1 Complete Blood Count (CBC)

A blood test generally examines the diverse characteristics of blood cells in a blood sample. Blood cancers may be identified using the blood test if numerous or very less type of blood cell or abnormal cells are noticed. A bone marrow biopsy may assist the diagnosis of leukemia [13].

1.1.6.2 Urine Cytology

Evaluating a urine sample under the microscope may showcase cancer cells that exist in the bladder, ureters or kidneys.

1.1.6.3 Blood Protein Testing

Electrophoresis is a test to examine various proteins in the blood. This can assist in discovering certain abnormal immune system proteins (immunoglobulins) that are
sometimes developed in patients with multiple myeloma. Other tests, like bone marrow biopsy are mainly done to confirm a suspected diagnosis [14].

1.1.6.4 Tumour Marker Tests

Tumour cells produce chemicals called tumour markers that can be found in the lineage. However, tumour markers are developed by some normal cells as well in the physical structure where the levels may be high in non-cancerous state. This defines the possibility for tumour marker tests in diagnosing cancer.

1.1.7 Molecular Basis of Cancer

Cancer is a composite genetic disease that may affect the future generations to come. The genetic injury may be obtained in somatic cells by environmental medium or acquired in the germ line. Tumours start to grow as clonal progeny of a single genetically affected progenitor cell. The monoclonality of tumours can be confirmed by study of X-linked markers (eg. Glucose 6-phosphate dehydrogenase iso-enzymes or X-linked restriction fragment length polymorphisms) [15].

Four factors are the targets of genetic harm.

- Growth promoting proto oncogenes
- Growth inhibiting tumour suppressor genes
- Genes that regulate apoptosis
- Genes that regulate DNA repair

The traits of malignancy (eg. Invasiveness and excessive growth which escapes from the immune system) are obtained in a stepwise method called tumour progression. At the genetic level, progression comes from collection of successive variations.

1.1.8 Treatment of Cancer

There are two common ways to tackle all characteristics of malignant neoplastic cancer. They are chemotherapy and radiation therapy. Apart from these two types, there are other methods of treatment available in current scenario. They are surgical procedure, biological and hormone therapy [16].
1.1.8.1 Chemotherapy

Chemotherapy is one of the most methods in the treatment of cancer patients. It is usually opted for patients whose cancer is metastasized. It helps to reduce the symptoms and pain. It also helps to slow down further development of tumours. At times chemotherapy also helps to destroy the cancerous cells [17].

Different combination of drugs are used in chemotherapy where the drugs are ingested through mouth or injected directly into the blood stream. Dosages are given to the patients in regular intervals for a period of time. The interval between each dosage is decided considering the type of cancer and the way the patient reacts to the medication. Drugs used in chemotherapy kills the cells that have a tendency to multiply fast in the body. In this process the healthy cells that multiply quickly also get destroyed by the drugs. Hair loss, nausea and fatigue associated with chemotherapy is caused due to this destructions of healthy cells. The side effects can be minimized to a certain level by combining other treatment methods and altering the frequency of the injection of chemotherapy drugs. Even though it is a long process, the side effects usually disappear once the course of treatment is completed [18].

1.1.8.2 Radiation Therapy

Radiation therapy is a way of treating cancer that uses radiations. It is generally effective in localized cancers. The radiation destroys the genetic material in the cancer cells and thereby reduces the power to multiply further. As in the case of chemotherapy, during radiation therapy too, the normal healthy cells get destroyed. It is found that once the process is completed, the healthy cells are regaining their genetic materials.

The ways by which the radiation therapy can be performed is internal and external. In the external radiation therapy with the help of special machines concentrated rays of radiation are given to the targeted portion of tissue in the body. In the internal radiation therapy radioactive elements are placed closed to cancerous tumours or the affected body areas by means of surgical procedure. Prior to insertion the origin of radiation is sealed in a tiny section like a catheter or capsule [19].
1.1.8.3 Surgery

Surgery is one of the very first methods used in the treatment of cancer. Today surgeries are no more a popular treatment option among the oncologists. Different cancer types need unique methods of operation. The cases of surgery include:

- Curative surgery is suggested when cancer cells are localized and they can be removed completely by an operation.
- Diagnostic and staging surgeries assist doctors to understand the extent, the cancer cells have spread.
- Surgery is done to help the medication process or lower the negative effects of treatment [20].

In most cases, surgery abolishes only a portion of the tumour, and radiation or chemotherapy must be carried out to kill the remaining cancer cells. This surgery is common during the times when removing the complete tumour may affect nearby organs.

1.1.8.4 Biological Therapy

In the biological therapy the body’s immune system is activated to attack the cancerous tumours. Monoclonal antibodies, interferon, and interleukin-2 play a vital role in biological therapy [21].

Side effects are found in biological therapy too, the intensity of which varies with the mode of treatment. Common symptoms are fever, loss of appetite, fatigue, bleeding, diarrhoea, muscle aches, rashes, swelling and vomiting [22].

1.1.8.5 Hormone Therapy

Hormone therapy averts cancer cells from utilizing the hormones they need to develop. The therapy may include the economic use of medicines that stop the production of certain hormones. Removing the hormone producing organs is also a part of treatment method.

Hormone therapy can have serious consequences including:

- Blood clots
- Significant changes in appetite
- Fluid retention
• Hot flashes
• Nausea
• Tiredness
• Weight gain

Some patients also face infertility problems later. The term and potency of the side effects are different according to the type of endocrine therapy.

1.2 MEDICINAL PLANTS

Biodiversity is gifted with a variety of medicinal plants that provides mankind with an unlimited source of treatment options. India is a tropical country with rich floristic constitution of medicinal plants. A huge percentage of population (80%) in the developing nations still relay on the herbal remedies for various diseases. They are in use for thousands of years and have been found beneficial in treating several diseases till date [23].

Medicinal plants are the primary sources of drugs for indigenous health care systems like Ayurveda, Unani and Siddha and for modern pharmacology as well. Some estimates show that, over 25,000 potential plant based drug treatments are available in Indian medicine, 1.5 million Specialists use medicinal plants in promotional and therapeutic usages [24]. Approximately 119 drugs are manufactured and marketed in Indian market from the 7500 medicinal drugs manufacturing units in India where 2000 stacks of medicinal herbs are consumed manually. Around 75% of these medicinal herbs used in the drug manufacturing units are part of the traditional herbal medicines widely used in the country. Over the years due to deforestation and over abuse of forests for various purposes, medicinal plant resources are deflated and there is a high alarm ringing for preservation of medicinal plant germplasm resources. By creating awareness on the significance of medicinal plants the preservation of resources has to be achieved [25].

Many of these medicinal plants are the rich sources of drugs that are extensively used to treat numerous disorders by the traditional practitioners. The medicinal plants were spotted and extracted for biochemical profile and conceived for medical applications. They acquire drugs importantly from wild plants and plants under culture. The drugs manufactured from the medicinal plants are used in treating high blood pressure, de-worming, counting diabetics, and as drugs to treat cancer [26].
1.2.1 Medicinal Plants as an Important Source of Drugs

Some of the methods employed to isolate compounds for drugs are synthetic chemistry and molecular modelling, despite the latest interest in molecular modelling. Even though new methods like combinatorial chemistry and other synthetic chemistry techniques are used by pharmaceutical companies, it has not reduced the demand for natural commodities, medicinal plants in particular. They still remain as a vital source for new drugs, fresh drug leads and raw chemical entities. On an average, every year four herbal medicines are being introduced into the US market which is an indicator to the growing popularity of the same [24].

1.2.2 Traditional Indian Systems of Medicine

The Indian subcontinent is gifted with a rich knowledge of health traditions that provide remedies for all the diseases as in the allopathic system of medication. The traditional medical system of India has two streams. The first is the local folk system followed in the rural and tribal villages. This system have been safeguarded and transferred through generations of housewives, birth attendants, skilled bone setters, the acupressure practioners, the vaidyas or doctors who treat snake bites, eye disorders etc. This health care system ensured everyone in the village to get medical assistance and care that gets extended to the state supported system which formed the next level of health care [27].

The second level in this native health system is the academic or classical system of Ayurveda, Siddha, Unani, Yoga, Naturopathy and Amchi. These systems of medicine have advanced foundations, both philosophical and theoretical codified in texts which are available in the form of manuscripts. The Indian Systems of Medicine (ISM) has therefore assimilated into it the systems of medicines that have originated in the subcontinent as well as other systems that have their birth place outside the land, Unani for instance.

Herbaceous plants have been the basis for the huge medical systems in human history, of Hippocrates and Galen, the great Islamic medical eras, and the Charaka and Susruta of the Indian sub-continent. Herbal drugs occupies an important place in all the traditional systems of medicine. Almost 3000 plants have been scientifically explored for their rewarding uses based on reference from traditional medicine. Interest in native medicines/drugs is not novel but has been encouraged in
recent years by methodological advances in phytochemistry, ethnobotanical studies, and of the awareness in renewable resources and conventional medicine[28].

The last decade has witnessed a renewed interest in the research of herbal medication in both developed and developing nations. Even before a couple of decades, Vaidyas used to prescribe customized mode of treatment to each patient, considering the body composition and nature of each one. With the introduction of technology, it became possible to produce medicines on a large scale in the manufacturing units or factories. Along with this, issues popped up like the unavailability of raw materials, availability of proper standardization methodology for the authentication of raw material, quality control parameters for single drugs and formulations etc., [29].

Chromatographic finger printing technique is the most remarkable methods that is used for the regular herbal drug analysis and for the quality assurance. As herbal medicines are of plant origin, they are susceptible to contamination, deterioration and variation in theme. This paves way to low quality of herbal products with minimal therapeutic efficiency. Quite regularly, the wanted biological response is due to not one ingredient but an array of bioactive components and the relative balance of important ingredients can vary from plant to plant of the similar species and in varied plant components. It is thus essential to establish internationally recognised guidelines for assessing quality. The role of traditional medicine in health care is acknowledged by World Health Organization (WHO) and has established the standard to be followed in the production of herbal medicines [30].

The World Health Organization – in its settlements, has imposed the requirement to pledge the quality of herbal medicine by applying modern control techniques and using suitable standards [29]. Hence, it is necessary to unfold methods for fast, accurate and specific recognition and estimation of active elements in order to deliver the consistency of vital elements in the manufacturing. Hence, before moving to clinical surveys, scientists need to set up a standard to verify plants and expose their strength [31].

1.3 FICUS SPECIES

The Ficus is a genus that has around 850 species of woody trees, bushes, vines, epiphytes and hemiepiphytes under the family Moraceae [32]. The Ficus are tropical
collection of plants and the best group in the United States. Ficus are the vital family contributing true trees for interior scraping. Integrating all the fig trees, they are genesis throughout the tropics of which very few species extend to the semi-warm temperate zone. Figs have transformed into over 850 varied species, of which the Ficus genus is the most crowded in count of species of all plant genera [33]. The genus Ficus belongs to the Mulberry family (Moraceae) and is the most prominent member of this family’s genera. It is present throughout the globe, finding its niche everywhere.

1.3.1 Fig Fruits

In Chinese, the fig is called as “no flower fruit,” which itself is a clue to its intricate composition [34]. It can be compared to pomegranates where there are a number of seeds, each a small fruit in itself. A plant of the moist tropics and subtropics, where it is found at elevations from 500 - 1,200 metres. The Ficus benghalensis fruits ripen annually twice. It grows best in areas where annual daytime temperatures are within the range 26 – 36 °C, but can tolerate 9 – 40 °C. The fruits are globose, sessile in axillary pairs, fleshy pericarp and with achene’s embedded in them, dark red in colour, 1.5-2.0 cm diameter, red to dark purple when ripe; seeds are tiny. The flowers (achenes) are concealed within the fig fruit and are not visible.

1.3.2 Ficus benghalensis

Ficus benghalensis commonly known as Banyan tree in India, Srilanka and Bangladesh, is found all over the country. It is also called as Bengal fig, Indian fig, East Indian fig, or Indian Banyan. It’s one among the four holy trees “Nalpamara” (Ksirivarkas) that are planted in the temples [35]. Their broad spread branches denote eternal life, and hence it is sacred. The banyan tree has various spiritual and mythological references. The unique character of the tree is that it has roots growing from its branches that grow downward towards the ground and turn into trees with their own rigid trunk. One Banyan tree gives an effect of a forest clustered with numerous trees. The original trunk roots along with the newer roots-turned-trunks support the wide spread tree. The average height of the Banyan tree is 21 m and lives for hundreds of years [36].

The Ficus benghalensis has notable large leaves and small flowers that turn into red fruits that resemble cherries. There is another fruit, which looks like a fig that grows on Banyan tree which is not edible. It acts like a container for a flower
holding structure called cycenia. The inside of the fruit has male and female flowers with males having pollen and female having seeds. Banyan tree has fragile and porous wood which is shown in Fig. 1.1. The tree generates sticky latex that the hunters use to create birdlime to capture birds.

![Fig. 1.1. Ficus benghalensis tree.](image)

The Indian banyan tree is revered in Indian culture and is considered as a holy or sacred tree. The *Ficus benghalensis* is very commonly seen in ancient temple complexes. The Indian Government has also published a postal stamp of the banyan tree to honour its contribution in social and historical times. Banyan trees are unique in their nature of growth as well. Birds lay the seeds into the top branches of other trees. The seeds sprout in the tree tops and the tree starts its journey as an epiphyte on host tree collecting its nutrition and water from the air and legs. The lateral branches grow the roots downwards to the soil. These supportive branches then grow into trunks and initiate new branches.

Apart from being an object of worship, Banyan is a sheltering tree with its shade that is extremely useful for travellers. It provides food for a number of birds
especially the Indian mynas which in turn facilitate the sprouting of new plants. One of the largest banyan tree in terms of area, is found in the Indian botanical garden of Howrah. It has found its place in the historical records that there was a banyan tree in the banks of river Narmada has won the admiration of Alexander the great. This tree covered huge area that it could shelter at least 7000 people. James Forbes in his oriental memories (1813-1815) has recorded that a banyan tree is nearly 2000 feet in circumference with 3000 tree trunks. The banyan tree in Calcutta botanical garden is more than a century old. Its main trunk is 13 feet in diameter with 230 trunks that are large as oak trees and more than 3000 smaller ones. The largest Banyan tree is in Sri Lankan Island. That tree boasts of 350 large trunks and more than 3000 large trunks and more than 3000 small ones [32].

1.3.3 Taxonomical Classification

Kingdom: Plantae
Division: Angiospermae
Class: Dicotyledonae
Order: Tubiflorae
Family: Moraceae
Genus: Ficus
Species: benghalensis

1.3.4 Distribution

The banyan tree is a native of South Asia, more specifically in India, Sri Lanka and Pakistan. It is often grown close to temples and is of spiritual significance. It is considered as a sacred tree by Hindus and Buddhists. It grows well in tropical, semi-tropical regions and monsoon and rain forests with mild to sufficient rainfall, humid air and wet soil. Its strongly drought resistant, withstands mild frost that is well suited for its growth. It is commonly found in the lower Himalayas in particular and all over India too.
1.3.5 Morphology

Banyan is an immense tree, with branches extending across large area; it grows to a height of 100 feet and, with its enormous limbs beared by prop roots, spread over several acres. The bark of the tree is thick and even. The colour ranges from greenish to white, former when young and later when mature. When cut to sheaths, it turns to pink. The wood is characterised by its softness and porous nature with milky and sticky latex.

1.3.6 Leaves

The leaves of the banyan tree are found near the end of the branches. They are shiny, leathery and wide-open towards maturity. They are mostly obtuse, base is circular, thickly rugged, basal nerves 3-7, the midrib with 4-6 pairs of secondary nerves, blade 10-20 cm, petiole 2-5 cm long, shoot pubescent, 1.5 to 2 cm in diameter, sessile, scarlet and red when ripe [37].

1.3.7 Flowers

The flowers of the banyan tree are tiny and has individual, male and female flowers. The male flowers throng near the lip of the receptacle. The female flowers are with shorter perianth and long style. The male and female flowers are found in the same receptacle [38].

1.3.8 Fruits

Fruits are globose and sessile in axillary pairs with fleshy pericarp and with achenes trenched in them and the colour is red, 1.5-2.0 cm in diameter, reddish to dark purple when mature. The seeds are small and is shown in Fig. 1.2. Fruits have cyanidin, rhamnoglycoside and polysaccharides. Yield is not for human consumption but edible to birds and monkeys [39].

Fig. 1.2. *Ficus benghalensis* fruits (figs)
1.3.9 Roots

Horizontal branches consistently produce aerial roots that grow downward into the soil to form new trunks. These widespread trunks prop up the tree and forms a huge horizontal mass. The tree species cannot be differentiated by the appearance of its prop roots. Older banyan trees are distinctive of various prop and buttress roots of large area [40, 41].

1.3.10 Chemical Composition

Stem bark is the house of numerous anthocyanidin derivatives (methyl ethers of Leucodelphinidin - 3 - O - L-rhamnoside, leucopelargonidin – 3 – O – L -rhamnoside. Lecocyanidin-3-O-D-galactosyl cellobioside) and aliphatic long chain ketones (pentatriacont-5-one, tetratriacont-20-en-2one, heptatriacont-6-en-10-one), besides-beta-sitosterol glucoside and mesoinsitol. Leucodelphinidin derivative, bengalenoside: Aglucoside, Leucopelargonin derivative, leucocynidin derivative, glycoside of leucopelargonidin have been detached from the bark of the Ficus benghalensis [42].

It is found that the leaves contain 9.63% of crude protein, 26.84% of crude fibres, 2.53% of calcium oxalate and 0.4% of phosphorous. Various chemical tests of ethanol extract and aqueous extract of leaves have identified flavonoids, sterols, tannins, phenol, and saponins in more quantity. There are no aromatic acids, gums, volatile oils, mucilage, carbohydrates and triterpenoids, in this plant. The flavonols of the leaves have been identified as quercetin-3-galactoside and rutin. Quercetin-3-galactoside, friedelin, rutin, lupeol, taraxosterol, β-amyrin along with psoralen, bergapten and β-sisterol are produced by the leaves [43].

1.3.11 Medicinal Uses

Various parts of the tree have medicinal properties. Milky juice and seeds of fruits are of immense use in outer application to aches, injuries, sores and ulcers, rheumatism and lumbago to the feet and to teeth and gums in odontalgia. Extract of fruits is smeared in cases of sukra roya of eye. Extract is useful in dysentery and diarrhoea. Infusion of bark is beneficial in lowering blood sugar in diabetes, dysentery, gonorrhoea and tonic. The bark concoction is productive as an astringent lotion in leucorrhoea. Leaves are boiled and spread as poultice to blisters and wounds to
enhance suppuration and elimination of pus. Leaves in decoction when used with roasted rice are considered as diaphoretic [44].

Root-fibers in form of mixture with or without addition of honey are similar to soft drinks in action—valuable in haemoptysis. Tender ends of aerial roots are an important dose to obstinate vomiting. Infusion of juvenile buds is used in cases of dysentery and diarrhoea [45]. Concentrated extract in combination with fruit was utilized as an aphrodisiac, in spermatorrhoea and gonorrhoea. Slender twigs of tree are collectively used as a good tooth brush, and it fortifies gums and teeth. It is found to be beneficial to women in treating various diseases of the reproductive system like amenorrhoea, menorrhoea, lemorhhoa, diminished lactation, etc. [46].

1.4 PHYTOCHEMICAL SCREENING

Extraction by natural means is based on the texture and liquid content of the plant under extraction and the case of subject matter that is being discussed upon. Generally, it is acceptable to homogenise the plant tissue, i.e. inhibiting enzyme oxidation or hydrolysis to happen and plucking new leaves, flower tissue and fruits suitably. If necessary boiling with ethanol is a safe method of reaching the goal. Alcohol in any case is a good multi-purpose solvent for initial extraction [47].

The authentic chemical method for getting organic constituents from dried plant tissue is to uninterruptedly extract, powdered contents in a soxhlet apparatus within a range of solvent, starting to turn with petroleum ether and chloroform and then utilizing alcohol and ethyl acetate. Yet, one unusually attains full separation of component parts and the same compounds may be derived in many fractions [48]. When researching the complete phytochemical profile of a plant species, fractionation of a crude extract is accepted in society to differentiate the master categories of components from each other, before chromatographic analysis. The quantities and type of compound isolates into the various fractions that may change from plant to plant.

The plant substances that have the common feature of an aromatic ring bearing one or more hydroxyl substituent are constituted under the broad term phenolic compounds. Phenolic substances are found in a cell vacuole and are water-soluble as they are commonly found combined with sugar as glycosides. The flavonoids form the biggest group, but simple monocyclic phenol, phenyl propanoids and phenolic quinines are also present in considerable numbers.
1.5 PHARMACOLOGICAL ACTIVITIES

Each biologically active compound has some of the pharmacological activities. Its concentration of action is comparative and is decided by the clear characteristics of object, dose, route, etc. On the other hand, the biological ability of compound includes all bodily functions that are under certain experimental conditions. Several medical properties have been attributed to Ficus benghalensis. Fruits of Ficus benghalensis is known to possess therapeutic activities and has been used by medical practitioners as an anti-diabetic, hypolipidemic, anti-inflammatory, anti-diarrhoeal, hepatoprotective, anti-asthmatic and anti-cancerous drug. Ficus benghalensis fruits are widely used in ayurvedic and sidhha treatments. The following section discusses its various therapeutic uses in medicine. The pharmacological activity explains the beneficial or adverse effects of a medicine on living things [49].

1.5.1 Antimicrobial Activity

Antibiotics are most important in fighting bacterial infections and have immensely gained from the health-related quality of human life since early times. Yet, these health benefits are under threat as many commonly used antibiotics have started having less fighting qualities against certain diseases, not just due to their toxic reactions, but due to the arrival of drug-resistant bacteria. It is essential to explore newer medicines with lower bacterial resistance [50]. Drugs obtained from natural sources are an important part in the prevention and handling of human illness. The inclusion of antibiotic resistance by the pathogenic microorganisms to conventional drugs has demanded the search for new, efficient and cost effective drugs to control infectious diseases. Various stories have specified that the medicinal plants possess a capital beginning for the isolation of active drugs for the control of infective organisms.

In an effort to cover the various nature of antimicrobial agents from natural resources. Ficus benghalensis has been found to be a useful antimicrobial agent against numerous bacterial and fungal diseases. Ficus benghalensis showcased vital antimicrobial activity and established properties that assist family medicine as broad-spectrum antimicrobial agents.
1.5.2 Antioxidant Activity

Antioxidant compounds in food have a vital part as a health protecting factor. Scientific proof reveals that antioxidants decrease the risk for chronic diseases including cancer and heart disease. Primary sources of naturally prevalent antioxidants are whole grains, fruits and vegetables. Plant sourced food antioxidants like vitamin C, vitamin E, carotenes, phenolic acids, phytate and phytoestrogens have been identified as possessing the future to decrease disease risk. Majority of the antioxidant compounds in a usual diet are obtained from plant sources and are a part of many classes of compounds with a broad variety of physical and chemical properties [51]. Some compounds, like gallates/salts, have powerful antioxidant activity, while others, such as the mono-phenols are fragile antioxidants.

The primary characteristic of an antioxidant is its capacity to trap free radicals. Highly reactive free radicals and oxygen species are in the biological systems from varied types of sources. These free radicals may oxidize nucleic acids, proteins, lipids or DNA and can start any of the degenerative diseases. Antioxidant compounds like phenolic acids, polyphenols and flavonoids scavenge free radicals such as peroxide, hydroperoxide or lipid peroxyl and thus stop the oxidative mechanisms that cause degenerative diseases. There are numerous clinical studies recommending that the antioxidants in fruits, vegetables, tea and red wine are the primary factors for the noted effectiveness of these foods in decreasing the happening of chronic diseases including heart disease and cancers. The free radical hunting activity of antioxidants in foods has been researched and documented [52].

1.5.3 Anticancer Activity

Ethnomedicinal plants, vegetables, fruits and herbs that are a part of folk and traditional medicine are now considered as one of the important sources of cancer chemoprevention drug discovery and development [53]. Complete 60% of the clinically utilized anticancer drugs are of natural origin and majority of them are obtained from higher plants. Considerable progress has been achieved in cancer chemotherapy; however, the concept of cancer medication using chemotherapy in various parts of the world has shifted considerably, based on the fact that, chemotherapy for cancer medication causes a major suffering to the patients. On the other hand, consuming full herbal preparations has produced less effect on the human
body due to the synergy of the plants that form the ingredients [54]. Moreover, ethnomedicinal plants for treating cancer are greatly cheap herbal drug treatment that can be economical for the rural and poor people to reach out efficiently several deep rooted cancers [55].

New anticancer agents are classified in varied categories based on their chemistry, bioactivity profile, and mechanism of activity [56-58]. Also, their clinical usage is usually given in combination with current therapy, using the fool-proof rules and profits of combination drug therapy to succeed clinical outcomes better than the then accessible medication [59-61]. Treating cells with the cytotoxic compound can lead to cell fates. The cells may experience necrosis, where they lose membrane integrity and fail fast because of cell lysis [62-64]. The cells can stop fiercely multiplying and dividing, or the cells can switch-on a genetic program of controlled cell destruction (apoptosis) [65-68].

1.6 CHROMATOGRAPHY

Generally the term chromatography originate from the native use for separating yellow and green plant pigments [69]. The chromatographic techniques has derived to separate mixture of compounds and to be considered as a very good separation technique.

The applications of chromatographic methods are

- The filtration of response blends in chemical synthesis,
- The filtration of bio-atoms, for example, proteins for pharmaceutical exploration,
- The investigation of complex example blends, for example, those got in criminology (body liquids, paints and so forth) and
- The investigation of ecological specimens [70].

In many cases, chromatography is used by researchers simply as a tool to get at a particular protein or other biological molecule.

1.6.1 Types of Chromatography

The differentiation is based on techniques of chromatography or principle or physical changes used.
They are

- The physical states of stationary phase and mobile phases.
- The principle of separation used.
- The chemical nature of stationary phase and mobile phases used (polarity).
- Based on the shape of stationary phase employed.
- Based on the purpose of chromatography experiment.
- Based on the physical or chemical characters of the stationary phase.

The above mentioned types of chromatography are theoretically defined. But practically there are only 11 types of chromatography [71].

1.6.1.1 Column Chromatography

Column chromatography in science is a strategy used to purge singular substance mixes from blends of mixes. It is regularly utilized for preparative applications on scales from micrograms up to kilograms. The principle point of preference of segment chromatography is the moderately minimal effort and additional of the stationary stage utilized as a part of the procedure [72].

1.6.1.2 High Performance Liquid Chromatography (HPLC)

High performance Liquid Chromatography (HPLC) is a type of section chromatography that pumps an example blend or analyte in a dissolvable (known as the versatile) stage at high weight through a segment with chromatographic pressing material (stationary stage). The example is conveyed by a moving transporter gas stream of helium or nitrogen. HPLC can isolate, and recognize extends that are available in any example that can be broken up in a fluid in follow focuses as low as parts per trillion. In view of this flexibility, HPLC is utilized as a part of an assortment of modern and experimental applications, for example, pharmaceutical, natural, legal sciences, and chemicals [73].

1.6.1.3 Gas Chromatography

It includes a specimen being vaporized and infused onto the head of the chromatographic segment. The specimen is transported through the section by stream of idle, vaporous versatile stage. The segment itself contains a fluid stationary stage which is adsorbed onto the surface of an inactive solid [74].
1.6.1.4 Ion-exchange Chromatography

Ion-exchange chromatography is a chromatography procedure that isolates particles and polar atoms taking into account their liking to the particle exchanger. It deals with any sort of charged atom—including extensive proteins, little nucleotides, and amino acids [75].

1.6.1.5 Size Exclusion Chromatography

The section is stacked with charged, gel having pores. Test particles when poured alongside versatile stage needs to go through the strainer like system of the stationary stage. In doing so, the bigger particles elute out first and slighter ones last. The reason is the slighter ones take longer way in the segment stationary stage while bigger particles take short way to elute out [76].

1.6.1.6 Thin Layer Chromatography (TLC)

Thin layer chromatography is a chromatography system used to unpredictable blends. It is a normally utilized strategy as part of manufactured science for distinguishing mixes, deciding their immaculateness and taking after the advancement of a response. It additionally allows the advancement of the dissolvable framework for a given partition. In examination with section chromatography, it just requires little amount of the compound and is much quicker too [77].

1.6.1.7 High Performance Thin Layer Chromatography (HTLC)

High Performance Thin Layer Chromatography is a chromatographic method that uses the fine activity of a dissolvable and a stationary stage to isolated mixes in a sample blend. A meager layer of adsorbent material is covered on a sheet of glass, aluminium foil or even plastic [78].

1.6.1.8 Paper Chromatography

In paper chromatography, the blend is spotted onto the paper, dried and the dissolvable is permitted to stream along the sheet by narrow fascination. As the dissolvable gradually travels through the paper, the diverse mixes of the blend separate into various shaded spots. The paper is dried and the position of various mixes is imagined. The guideline behind the paper chromatography is that the most solvent substances move
further on the channel paper than the minimum dissolvable substances. Distinctive plant colours can be isolated by utilizing the strategy of paper chromatography [79].

1.6.1.9 Affinity Chromatography.

It is the favoured strategy for bio-selective adsorption and consequent recuperation of a compound from an immobilized ligand. Each is intended for profoundly particular and proficient decontamination of proteins and related mixes [80].

1.6.1.10 Liquid Chromatography–Mass Spectrometry (LC-MS)

It is a logical science research laboratory strategy for recognizable proof, quantitation and mass examination of materials. This method takes into account the auxiliary illustration of obscure particles through fracture. Like HPLC, LC/MS uses a compound's inborn fondness for both a "portable stage" (normally a cradled dissolvable) and a "stationary stage" (permeable strong backing with particular covering) [81].

Basically, a pump is utilized to give a constant stream of a dissolvable into which a broke up test is presented. Once the example is in the dissolvable stream, it goes through a systematic segment. The mixes present in the specimen blend are then isolated relying upon their fondness to the covered particles in the segment. After the segments in the specimen are isolated, they go through a mass identifier. The mass finder reaction and the "maintenance time" (time it takes for a compound to go from the injector to the identifier) of the compound(s) of interest may then be contrasted with a reference material.

1.6.1.11 Gas Chromatography Mass Spectrometry (GC-MS)

It utilizes a compound's intrinsic affinity for a "stationary phase" (solid support with specialized coating) and facilitates the separation of complex sample matrices into their component parts [82].

1.6.1.12 Ultra High Performance Liquid Chromatography

Critical advances in instrumentation and section innovation were made to accomplish emotional expansions in determination, pace and affectability in fluid chromatography. Surprisingly, an all-encompassing methodology including
concurrent advancements in molecule innovation and instrument configuration was tried to meet and conquer the difficulties of the systematic research centre [83].

1.7 NANOTECHNOLOGY AND NANOMEDICINE
1.7.1 Nanotechnology

Nanotechnology has multiple definitions but commonly it is the use and execution of materials with sizes in the nanometre range. A millimetre is one-thousandth of a metre, similarly, a nanometre is one-millionth of a millimetre. To be clear, a human hair is approximately 80,000 nanometres in diameter and the expanding science and industry of nanotechnology uses materials lower than 1000 nanometres. Advantage of working at the nano scale have been seen in different places as electronics and energy storage to sunscreens and food packaging [84].

1.7.2 Nanomedicine

Nanomedicine is simply the practice of nanotechnologies in a healthcare setting and the vast benefits that have been noticed include the use of nanoparticles to better the behaviour of drug substances. Nanomedicines, nowadays are used worldwide to enhance the treatments and lives of patients having numerous disorders like ovarian and breast cancer, kidney disease, fungal infections, elevated cholesterol, menopausal symptoms, multiple sclerosis, chronic pain, asthma and emphysema. The nanomedicines currently in handy are overpowering the hardships experienced by normal medical treatments in providing the aid from the drug molecules utilized. Sometimes the drugs have very less solubility in water and the human body endeavours to absorb the sufficient amount to treat the prevailing condition [85]. Otherwise, the drug molecule is digested well, however the body eradicates the medicine before it can provide a benefit. Drugs may lead to negative consequences due to poor delivery at the actual site of disease. For instance, medicines that are targeting cancers must be away from healthy tissues and organs or damage can be caused. Nanomedicines can hence have a vital role in confirming sufficient drug enters the body, it is retained in the body for long periods and is targeted mainly to the portions that are in need of treatment [86].

It's known for few years that recognizing the disease in early stages can help prevent prolonged damage or even death. It is much common that many diseases that have no symptoms are seen for numerous years but the human body definitely
generates evidence of disease at the molecular level. Another vital area of nanotechnology and nanomedicine is diagnostics [87].

1.7.3 Nanoparticles

A nanoparticle is a microscopic particle with at least one dimension less than 100 nm. Nanoparticle research is a branch of deep scientific study, because of huge scope of potential applications in biomedical, optical, and electronic branches of study. Nanoparticles are of huge scientific attention as they are actively a link between bulk materials and atomic or molecular structures [88]. A bulk material has steady physical properties nevertheless of its size, but that’s not the case in nano-scale. Size-conditional properties are seen such as quantum confinement in semiconductor particles, surface plasmon resonance in some metal particles and superparamagnetism in magnetic materials. The characteristics of materials differ as their size near the nanoscale and as the percentage of atoms at the peripheral of a material becomes significant. For bulk materials more than one micrometre the percentage of atoms on the surface is minuscule relative to the total number of atoms of the material.

Nanoparticles showcase numerous special properties relative to bulk material. Deferment of nanoparticles are possible as the intercommunication of the particle surface with the solvent is powerful to thrash the differences in density that results in a material either sinking or floating in a liquid. Nanoparticles often have unanticipated visible properties as they are tiny enough to confine their electrons and generate quantum effects.

1.7.3.1 Metallic Nano Particles

Metallic nanoparticles have conceivable applications in assorted ranges, for example, gadgets, beautifying agents, coatings, bundling, and biotechnology. For instance, nanoparticles can be prompted to combine into a strong at moderately bring down temperatures, regularly without liquefying, prompting enhanced and simple to-make coatings for hardware applications (eg, capacitors). Regularly, nanoparticles have a wavelength beneath the basic wavelength of light. This renders them straightforward, a property that makes them exceptionally valuable for applications in beautifiers, coatings, and bundling. Metallic nanoparticles can be appended to single strands of DNA non-destructively. This opens up roads for medicinal symptomatic applications. [89, 90].
Metal oxides assume an extremely huge part in material science for case manufacture of microelectronic circuits, sensors, piezoelectric gadgets, energy components, and coatings for the passivation of surfaces against consumption and as impetus [91]. Metal oxides have likewise been utilized as sorbents for natural toxin. In the area of nanotechnology, oxide nanoparticles can display exceptional compound properties inferable from their restricted size and high thickness of corner or edge surface locales. Various physical and synthetic preparative techniques for getting to nanostructured oxides are on records [92].

1.7.4 Nanoparticles Synthesis

Development of nanoscience and nanotechnology involves three key components viz material synthesis, characterization, and application. Synthesis and processing of nanostructures is based on bottom-up or top-down approaches, i.e. either to assemble atoms together or to dis-assemble bulk solids into finer pieces until they are composed of only a few atoms [93]. Both approaches are widely used and have their own advantages and disadvantages. Top-down methods such as attrition and etching are simple, fast, and have good scalability. However, crystal imperfections or impurities may arise in these techniques. Bottom-up methods are more preferred for obtaining highly pure nanomaterials with homogenous chemical composition and better short or long range ordering. Some examples include self-assembly, deposition techniques, epitaxial growth etc. Different synthesis routes result in different material properties such as crystallinity, microstructure, and chemical composition. Another way of classifying various synthesis methods is based on its nature: either physical, chemical, or biological. Physical approach may be mechanical, such as ball milling and melt mixing; or vapour based, such as sputtering, laser ablation, physical vapour deposition etc. These methods are free of solvent contamination and produce nanoparticles with uniform size distribution. Chemical methods like chemical reduction, microemulsion technique, photoreduction, electrochemical technique etc. are simple, enable low temperature synthesis, and can be scaled up easily [94].

A variety of shapes and sizes are also possible with this approach. A drawback, however, is that it involves toxic organic or inorganic solvents. The third approach, biological, tends to overcome this drawback by providing more eco-friendly methods. Also called as green synthesis, these techniques exploit various protocols of
nature to synthesize nanomaterials. It includes use of microorganisms, either eukaryotes (yeast, fungi) or prokaryotes (bacteria or actinomycetes), use of plant extracts or enzymes, and use of templates made of DNA, membranes, viruses or diatoms. Highly stable and well-characterized nanoparticles have been obtained from these methods by optimizing critical synthesis parameters [95].

1.7.5 Nano Characterization

Nanotechnology is expected to play a key role within the next 10 years in a wide spectrum of industry sectors including manufacturing, information technology, electronics, and healthcare. Novel devices at the micro- and nanoscale will become increasingly important in all of these industries [96]. The ability to measure dimensions, characterize materials, and elucidate structures of new and novel materials at the nanoscale will be critical to the advancement of nanotechnology. In addition, one of the exciting prospects of nanotechnology lies in the ability of molecules or particles, under specific conditions, to self-assemble and form new materials with unusual properties. Successful development of these new materials will require the ability to monitor such processes at the nanoscale in real time. Metrology, the science of measurement, is therefore the foundation of nanotechnology. Standards and reference materials will also provide essential infrastructural support to this emerging technology.

Prior to applications, the synthesized material has to be analyzed for its properties [97]. At the nanoscale, material properties significantly differ from that of bulk. For instance, mechanically it becomes stronger, high surface-volume ratio increases chemical reactivity, it has lower melting point, band-gap energy increases, magnetic properties are enhanced, and it exhibits blue shift due to the surface plasmon resonance effect. Quantum confinement also introduces a variety of electronic properties such as ballistic conduction and tunneling effects. All of these properties are highly dependent on the morphology and size of the particles. Therefore for effective applications, deeper knowledge of the potential of the synthesized particles is necessary.

Characterization is done by a variety of techniques, mainly derived from material science. It involves two main types: structure analysis and property measurement. Structure analysis includes determination of particle size, shape, surface
topography, pores, defects, microstructure etc. It is performed using a variety of microscopic and spectrometric methods like electron microscopy, scanning probe microscopy, X-ray diffraction spectrometry, etc. Characterizing the properties is rather difficult since nanomaterials show diverse properties which are highly selective to shape and size. Chemical characterization involves determining the surface and interior atoms and their spatial distributions. It includes optical and electronic spectroscopy, and ion spectrometry. Prominent examples are IR spectroscopy, X-ray spectroscopy, Auger electron spectroscopy, Rutherford backscattering spectrometry etc. Characterization of physical properties involves determining mechanical, thermal, optical, electrical, electrochemical, and magnetic properties. This is commonly done using atomic force microscopy, Thermo Gravimetry – Differential Thermal Analysis, electrochemical workstation, spectrometers, and other analytical instruments.

1.7.6 Bioconjugation

Bioconjugation is the binding of two or more molecules to form a novel complex having the combined properties of its individual components. Natural or synthetic compounds with their custom activities can be chemically integrated to form a unique substance having cautiously engineered characteristics. Thus, a protein that can bind distinctly to a target molecule in a complex mixture can be cross-linked with another molecule able to be detected to form a traceable conjugate. The detection component provides visibility for the targeting component, generating a complex that can be localized, followed via many processes, or used for measurement [98].

The technology of bioconjugation has influenced almost every part in life sciences. Application of the accessible crosslinking reactions and reagent systems for forming novel conjugates with distinct activities has paved way for the assay of minute quantities of substances, the in vivo targeting of molecules, and the modulation of custom biological processes. Modified molecules have been used for purification, for recognition of specific cellular components, and in the therapy of disease.

The possibility to chemically connect one molecule to another has lead to the birth of billion-dollar industries serving research, diagnostics, and therapeutic markets. A noteworthy part of all biological assays, including clinical testing, is now done with unique conjugates that can interact with particular analytes in solutions, cells, or tissues. Crosslinking and modifying agents can be used in altering the native
state and function of peptides and proteins, sugars and polysaccharides, nucleic acids and oligonucleotides, lipids, and almost any other conceivable molecule that can be chemically obtained.

1.7.6.1 Bioconjugation Applications

Bioconjugation techniques strives to apprehend the spirit of this field via three main sections: its chemistry, reagent systems, and principal applications. Bioconjugate Chemistry, starts with a going-through of the major chemical groups on target molecules that can be used in modification or crosslinking reactions. Bioconjugate applications debates on the preparation of unique conjugates and labelled molecules for use in distinctive application sectors. They are

- Preparing hapten–carrier conjugates for immunization, antibody production, or vaccine research;
- Manufacturing antibody–enzyme conjugates for use in enzyme immunoassay systems;
- Preparing antibody–toxin conjugates for use as targeted therapeutic agents;
- Making lipid and liposome conjugates and derivatives;
- Producing conjugates of avidin or streptavidin for use in avidin–biotin assays;
- Labelling molecules with colloidal gold for sensitive detection purposes;
- Producing polymer conjugates with PEG or dextran to modulate bioactivity or stability of macromolecules;
- Enzyme modification and conjugation strategies; and
- Nucleic acid and oligonucleotide conjugation techniques.

Each and every application area demands cutting-edge technologies that depend mainly on bioconjugation techniques. In most cases, without the possibility to connect one molecule to another most of the research advancement in these fields would halt. Nanoscaled systems for systemic cancer therapy and their latest stage of development are summarized in Table 1.1.
<table>
<thead>
<tr>
<th>S.No</th>
<th>Vehicle platform</th>
<th>Stage of recent development</th>
<th>Examples</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Liposomes</td>
<td>Approved</td>
<td>DaunoXome, Doxil</td>
</tr>
<tr>
<td>2</td>
<td>Albumin-based particles</td>
<td>Approved</td>
<td>Abraxane</td>
</tr>
<tr>
<td>3</td>
<td>PEGylated proteins</td>
<td>Approved</td>
<td>Oncospar, PEG-Intron, PEGASYS, Neulasta</td>
</tr>
<tr>
<td>4</td>
<td>Biodegradable polymer–drug composites</td>
<td>Clinical trials</td>
<td>Doxorubicin Transdrug</td>
</tr>
<tr>
<td>5</td>
<td>Polymeric micelles</td>
<td>Clinical trials</td>
<td>Genexol-PM, P1049C, NK911, NK012, NK105, NC-6004</td>
</tr>
<tr>
<td>6</td>
<td>Polymer–drug conjugate-based particle</td>
<td>Clinical trials</td>
<td>XYOTAX (CT-2103), CT-2106, IT-101, AP5280, AP5346, FCE28068 (PK1), FCE28069 (PK2), PNU166148, PNU166945, MAG-CPT, DE-310, Pegamotecan, NKTR-102, EZN-2208</td>
</tr>
<tr>
<td>7</td>
<td>Dendrimers</td>
<td>Preclinical</td>
<td>Polyamidoamine (PAMAM)</td>
</tr>
<tr>
<td>8</td>
<td>Inorganic or other solid particles</td>
<td>Preclinical</td>
<td>Carbon nanotubes, silica particles, gold particles (CYT-6091)</td>
</tr>
</tbody>
</table>
1.8 MOLECULAR DOCKING

Molecular docking is the much repeatedly used methods in structure-based drug design, due to its capacity to forecast the binding-conformation of small molecule ligands to the appropriate target binding site. Characterisation of the binding conduct has a vital role in rational design of drugs and also explain fundamental biochemical processes [99].

1.8.1 Ligands

A ligand is an ion or molecule that attach to a central metal atom to create a complex. Ligands are atoms or molecules with electron pairs in handy; they may be neutral or negatively charged. Ligands are considered as electron donors attracted to the metal (the electron acceptor) at the center of the complex. The ligand supplies both of the electrons for the bond that creates between itself and the central metal atom or ion. There are various types of ligands based on their binding potential [100]. They are

1.8.1.1 Monodentate Ligands

Monodentate ligands possess only one atom with the capacity of binding to a central metal atom or ion.

1.8.1.2 Polyydentate Ligands

In a polyydentate ligand the ligand molecule has more than one donor atom. They are given distinct names, determining by how many donor atoms they contain.

1.8.1.3 Bidentate Ligands

Bidentate ligands possess two atoms with the possibility of binding to a central metal atom or ion.

1.8.1.4 Tridentate Ligands and Higher

Tridentate ligands possess three atoms with the capacity of binding to a central metal atom or ion. Molecules possessing four donor atoms are known as tetradeate ligands; five donor atoms, pentadentate; and six donor atoms hexadentate. A complex that has a polyydentate is called a chelate.
1.8.1.5 Ambidentate Ligands

Ambidentate ligands are monodentate ligands that have the possibility to bind in two possible places.

1.8.2 Receptors

Receptors are protein-molecules that collects chemical-signals from outside a cell. When such chemical-signals attach to a receptor, they create a cellular/tissue-response. Here, a receptor is a protein-molecule that identifies and reacts to endogenous-chemical signals. Receptor-proteins are implanted in all cells' plasmatic-membranes; facing extracellular-(cell surface receptors), cytoplasmic (cytoplasmic-receptors), or in the nucleus (nuclear receptors). A molecule that attaches to a receptor is known as ligand, and could be a peptide (short-protein) or a small molecule like a neurotransmitter, hormone, pharmaceutical-drug, toxin, or parts of the outside of a virus or microbe. The innermost designated-molecule for a specific receptor is known as its endogenous-ligand. Membrane-impermeant signalling molecules can attach to and activate either [101]

- Channel-linked receptors
- Enzyme-linked receptors
- G-protein-coupled receptors
- Membrane permeant signalling molecules activate intracellular receptors.

1.8.3 Ligand Receptor Docking

Docking is a process that envisions the preferred alignment of one molecule to a second when attached to each other to form a stable complex. Knowledge of the preferred orientation can be helpful to envision the binding affinity between two molecules using, scoring functions. The associations between biologically relevant molecules like proteins, nucleic acids, carbohydrates, and lipids play a vital role in signal transduction. Also, the relative alignment of the two interacting partners may influence the type of signal generated. Hence, docking is useful for envisioning the strength and kind of signal produced [102].

Molecular docking may be considered as a hindrance of “lock-and-key”, where one wants to identify the right relative orientation of the “key” that can open the “lock”. Here, the protein can be considered as the “lock” and the ligand as a “key”.

33
Molecular docking is defined as a development problem that describes the “best-fit” alignment of a ligand that binds to a specific protein of interest. However, since both the ligand and the protein are pliable, a “hand-in-glove” analogy is more suitable than “lock-and-key”. During the docking process, the ligand and the protein regulate their conformation to reach an overall "best-fit" and this conformational adjustment consequencing in the overall attachment is known as "induced-fit".

1.9 DRUG DELIVERY

Drug delivery is a process of issuing a pharmaceutical compound to attain a therapeutic effect in humans or animals. For the medication of human diseases, nasal and pulmonary routes of drug delivery are gaining importance. These courses give confirmed alternatives to parenteral drug delivery specifically for peptide and protein therapeutics. For this, numerous drug delivery systems have been devised and are being scrutinized for nasal and pulmonary delivery. They comprise liposomes, proliposomes, microspheres, gels, prodrugs, cyclodextrins. Nanoparticles that include biodegradable polymers show commitment in fulfilling the stringent prerequisites placed on these delivery systems, like the capacity to be carried into an aerosol, stability against forces generated during aerosolization, biocompatibility, targeting of distinct sites or cell populations in the lung, release of the drug in a preset way, and wearing down within a period of time [103].

Development of new drug molecule is costly and prolonged. Improving safety efficacy ratio of “old” drugs has been strived using various methods like, individualizing drug therapy, dose titration, and therapeutic drug monitoring. Delivering drug at regulated rate, slow delivery and targeted delivery are other very fascinating methods and are taken-up rapidly [104].

1.9.1 Mode of Drug Delivery

Nanoparticles and Nano formulations have been implemented as drug delivery systems with huge success; and Nano particulate drug delivery systems have still greater prospects for various applications, including anti-tumour therapy, gene therapy, and AIDS therapy, radiotherapy, in the delivery of proteins, antibiotics, virostatics, and vaccines and as vesicles to pass the blood-brain barrier. Drug delivery technologies alter drug release profile, absorption, distribution and elimination for the benefit of improving product potency and safety, as well as patient convenience and assent. Drug
release is from: diffusion, degradation, swelling, and affinity-based mechanisms. There are various methods of drug administration. They are.

- Oral Administration
- Buccal/Sublingual
- Rectal, Intravenous (IV)
- Subcutaneous
- Intramuscular
- Inhalers
- Transdermal

1.9.2 Pharmacokinetics

Pharmacokinetics, is the travel of drug into, through, and out of the body—the time course of its absorption, bioavailability, metabolism, and excretion. Pharmacodynamics is outlined as what a drug does to the body, comprises receptor binding, post-receptor effects, and chemical interactions. Drug pharmacokinetics decides the onset, duration, and intensity of a drug’s effect. Formulas combining these processes summarize the pharmacokinetic behaviour of most drugs [105].

1.9.2.1 Drug Absorption

Drug absorption is ascertained by the drug’s physicochemical properties, establishment, and route of administration. Dosage forms (tablets, capsules, solutions), comprising of the drug and other ingredients, are setup to be given by different ways (oral, buccal, sublingual, rectal, parenteral, topical, inhalational). Drugs must be in solution state to be absorbed. Thus, solid forms (eg, tablets) must be able to disintegrate [106].

1.9.2.2 Drug Bioavailability

Bioavailability allude to the rate at which the active moiety (drug or metabolite) enters systemic circulation, thereby approaching the site of action. Bioavailability of a drug is mainly decided by the properties of the dosage form, which rely partly on its design and manufacture. Differences in bioavailability among compositions of a given drug can have clinical importance; thus, understanding whether drug formulations are equivalent is necessary.
1.9.2.3 Drug Distribution

After a drug enters the regulatory circulation, it is distributed to the body’s tissues. Distribution is usually not regulated due to differences in blood perfusion, tissue binding, regional pH, and absorbent nature of cell membranes. The entry rate of a drug into a tissue relies on the rate of blood flow to the tissue, tissue mass, and divisional characteristics between blood and tissue. Distribution equilibrium amidst blood and tissue is reached much faster in richly vascularized areas, unless diffusion across cell membranes is the rate-limiting step. After equilibrium, drug concentrations in tissues and in extracellular fluids are displayed by the plasma concentration. Metabolism and excretion happen at the same time with distribution, turning the process dynamic and complex [107].

1.9.2.4 Drug Metabolism

The liver is the vital site of drug metabolism. Though metabolism generally inactivates drugs, some drug metabolites are clinically active—sometimes even more than the parent compound. A weakly active substance that possess an active metabolite is a prodrug, especially if outlined to deliver the active moiety more effectively.

Drugs can be metabolized by oxidation, reduction, hydrolysis, hydration, conjugation, condensation, or isomerization. The enzymes taking part in metabolism are available in many tissues however generally are more concentrated in the liver. Some patients metabolize a drug so fast that clinically effective blood and tissue concentrations are not attained; in others, metabolism may be so slow that usual doses have toxic effects. Custom drug metabolism rates are altered by genetic factors, coexisting disorders and drug interactions.

1.9.2.5 Drug Excretion

The kidneys are the vital organs for expelling water-soluble substances. The biliary system supplies to excretion to the extent that drug is not reabsorbed from the GI tract. Usually, the contribution of intestine, saliva, sweat, breast milk, and lungs to excretion is small, except for exhalation of volatile anaesthetics. Hepatic metabolism usually rises drug polarity and water solubility. The resulting metabolites are then more promptly excreted.
1.9.3 Pharmacodynamics

Pharmacodynamics, explained as what a drug does to the body, comprises receptor binding (including receptor sensitivity), post receptor effects, and chemical interactions. Pharmacodynamics, with pharmacokinetics (what the body does to a drug), assists explaining the relationship between the dose and response. The pharmacologic response relies on the drug binding to its target. The concentration of the drug at the receptor site impacts the drug’s effect [108, 109].

A drug’s pharmacodynamics can be impacted by physiologic alterations due to disorders, aging, or other drugs. Disorders that influence pharmacodynamics responses comprise genetic mutations, thyrotoxicosis, malnutrition, myasthenia gravis, Parkinson disease, and some forms of insulin-resistant diabetes mellitus. These disorders can alter receptor binding, change the level of binding proteins, or lesser the receptor sensitivity. Aging tends to affect pharmacodynamics responses via changes in receptor binding or in post-receptor response sensitivity. Pharmacodynamics drug–drug interactions result in competition for receptor binding sites or alter post-receptor response.

1.9.4 In vitro Studies

In vitro studies are conducted with microorganisms, cells or biological molecules outside their biological background [110]. Colloquially called "test tube experiments", these studies in biology and its sub-disciplines have authentically been carried out in test-tubes, flasks, petri dishes etc. and since the onset of molecular biology demand techniques called omics [111]. Studies performed using components of an organism that have been isolated from their usual biological surroundings permit a more detailed or more convenient analysis than can be performed with whole organisms [112]. In contrast, in vivo studies are done in animals including humans, and whole plants [113].

The present study comprises the anticancer activity of metal oxide nanoparticles such as iron and zinc oxide nanoparticles bioconjugated with the extracts of fig fruit against various cancer cell lines. The novelty of this work lies with the extraction of fig fruits and determining the phytochemical compounds which act as a triggering agent for anticancer activity.