GENERAL INTRODUCTION

Since prehistoric time, alleviation of diseases has been one of the primary concerns of mankind. Local practitioners have used indigenous plants and herbs for centuries all over the world in a variety of ailments showing definitive pharmacological activities and even as a poison. Those plants, which provide toxic effect, were being used in hunting or warfare, while plant derived products like opium and hashish has long been used as hallucinating agents.

Traditional medicines developed over the millennia through the acquired experience and accumulated knowledge of man for the beneficial or harmful effects of plant material against various human sufferings, culminating in self contained theories. Thus man succeeded in discovering cure of ailments with herbal, animal and mineral medicines. Much indigenous medicine began as myth, transferred to new generation as folk medicine and developed with times as the complex science of modern medications.

The history of medicine is an account of man's effort to deal with human illness and diseases from primitive attempts of preliterate man to the present complex array of specialities in treatments and can be broadly into following eras. The early period comprising Indian, Chinese, Sumarian, Egyptian, Assyrian, and Babylonian civilization followed by Greek, Roman, Persian, Eastern and Medieval era and finally the modern period.¹ ²

The earliest traces of therapeutic use of plant are recorded in the Rigveda (4500-1600 BC) and Ayurveda (2500-600 BC). Charaka cited about fifty groups of herbs while Sushruta explained 760 herbs in 37 sets. Buddhists period boosted the use of medicinal plants and gave a considerable attention to cultivate these plants in scientific manner.³
The seeds of Chinese medicine were shown by Fu His (2953 BC) and were flourished under the rule of Chang dynasty. The emperor Chen Nung himself compiled a Pharmacopoeia called “Pen Tsao” on medicinal plants around 2735 BC. It consists of forty volumes with thousand prescriptions in which he reported antifebrile effects of drug “Chang Sheng” (*Dichroa febrifuga*) and stimulatory and diaphoretic effects of the drug “Ma Huang” from the *Ephedra sinica*. These drugs are still derived from this plant. Ben Cao Gang Mu was written by Li Shizhen and still serves as a valuable reference for teaching, practicing and guidance for medicinal research in China. Most of the Chinese medicine literature is found in “Nei Ching”.

The most civilization is supposed to be Egyptian, which have delineated the use of many medicinal plants for the treatment of human ailments in “Emerged Papyrus” as early as 1500 BC. The Assyrian and Babylonian pharmacy emerged during about 650 BC. Hippocrates (460 BC), the father of medicine, laid the foundation of pharmacy. He described and named early 400 samples as medicines. Theophrastus (370-267 BC) entered 500 plants as medicine in “On the History of Plant”. He received the garden herb of Aristotle as a legacy. He also wrote “On the Classes of Plants” and “Pharmacologic” handbook for approximately 1600 years. Dioscorides (60 AD) gave one of the most authentic and conclusive compilation of the Greek medicines. Then Pliny, the elder (23-79 AD) inscribed “Natural History” in 37 volumes.

Galen kept his pharmacy in Rome and prescribed so many preparations of plant origin that these preparations are called Galenicals. He is credited of 30 books in pharmacology.
With the advent of Islam, Arabs acquired, collected, translated and documented the knowledge of use of herbal medicine from almost all the famous civilizations. There is a long list of famous names of Arab scholars such as Abu Bakr Mohammed Bin Zakaria Al-Razi (Rhazes) who wrote nearly 250 remarkable works. A large part of his work on pharmacological subject is described in “Alhavi Kabeer” or “Continens of Rhazes”. His noticeable book “Kitab-al-Mansoori” exhaustively debates the Greek-Arab system of medicine. Beside his written which he was first to use opium as ‘anesthetic’ Ali Ibn-e-Al-Rabban Al-Tabari (833-870 AD) wrote the “Firdous Al-Hikmat” which comprises seven parts. The most famous physician, philosopher, mathematician and astronomer of his time, Bu-Ali Ibn-e-Sina, “Avicenna” (908-1037 AD) has documented and therapeutic uses in entailed 760 herbal drugs in his famous book “Qanun fi Al-Tibb” or ‘The Cannon of Medicine’. It was considered to be the most authentic Materia Medica and served as a standard textbook of medicine in Europe till the 17th Century AD. Ibn Al-Baiter, a great botanist and pharmacist has described a large number of medicinal plants and therapeutic uses in his book Kitab Al-Jami fi Al-Adwiy Al-Mufrada. His second monumental work Kitab Al-Mughni fi-Al-Mufrada is an outstanding encyclopedia of herbal medicine with twenty chapters, in which drugs are listed therapeutic. Al-Idrisi (1099-1166 AD) another well known scientist also made a significant contribution in medicinal plant therapy and has described his work in “Kitab Al Jami-ul-sifat Ashat Al-Nabatat”.

It is well recognized that modern medicinal science has its roots in the Islamic civilization of Middle Ages and resulted in the flowering of medical technology in Europe during the days of renaissance.
Almost two hundred years ago, about 1806 AD a new door for search of useful drugs. From the plant kingdom was opened with the isolation of morphine from dried latex of *Papaver sominiferum* L. (opium), by F. W. Sertturner (1783-1810). The isolation, physiological and pharmacological role of strychnine, papaverine, quinine, nicotine and cocaine isolated by Pelletier and Cavantou lead to more concentrated attention towards the chemical investigation of medicinal plants. The structure of cocaine was establishment by Hofmann in 1884 and was synthesized by Ladenburg in 1886. Due to the chemical complexity and their physiological role these compounds must be as the first of the typical natural products, isolated by the man as a pure compounds. A number of important discoveries in medicine can be attributed to the isolation of active principles from natural products.

Increasing use of herbal drugs and gradual collection of knowledge gave an impetus to establish Ayurvedic and Unani Tibbi College by Hakim Ajmal Khan at Dehli in 1920 and Hamdard University by Hakeem Saeed near Karachi in early 1990’s. Although the plants have served through the ages as the mainstay for the treatments of various ailments, it is astonishing that from the plant kingdom which comprises approximately 600,000 plant species only two percent of these species have been subjected to pharmaco-chemical studies.

Through early decades of the last century, the development of chemotherapy led to the increasing shift towards synthetic drugs. With the discovery of antibiotics, e.g, Penicillin by Alexander Fleming and other medicinal important constituents of medicinal plants, there has been a considerable recovery of interest in the study of natural product chemistry. According to WHO estimation more than half of the world population still
depends on the traditional medicine because of their ready availability, cheapness and socio-cultural background.

**Natural Products: In Modern Medicine**

The use of natural products with therapeutic properties is as ancient as human civilisation and, for a long time, mineral, plant and animal products were the main sources of drugs. The Industrial Revolution and the development of organic chemistry resulted in a preference for synthetic products for pharmacological treatment. The reasons for this were that pure compounds were easily obtained, structural modifications to produce potentially more active and safer drugs could be easily performed and the economic power of the pharmaceutical companies was increasing. Furthermore, throughout the development of human culture, the use of natural products has had magical-religious significance and different points of view regarding the concepts of health and disease existed within each culture. Obviously, this approach was against the new modus vivendi of the industrialized western societies, in which drugs from natural resources were considered either an option for poorly educated or low income people or simply as religious superstition of no pharmacological value. However, even if we only consider the impact of the discovery of the penicillin, obtained from micro-organisms, on the development of anti-infection therapy, the importance of natural products is clearly enormous. About 25% of the drugs prescribed worldwide come from plants, 121 such active compounds being in current use. Of the 252 drugs considered as basic and essential by the World Health Organisation (WHO), 11% are exclusively of plant origin and a significant number are synthetic drugs obtained from natural precursors.

Natural products have been investigated and utilized to alleviate disease since
early human history. In the early 1900s, before the “Synthetic Era”, 80% of all medicines were obtained from roots, barks and leaves. At that time, fluid extracts were in vogue. Trustful humanity placed its faith in the belief that for every ill there existed a cure in the plants of field and forest. As Rudyard Kipling wrote (1910), “anything green that grew out of the mould was an excellent herb to our fathers of old.” In more recent times, natural products have continued to be significant sources of drugs and leads. Their dominant role is evident in the approximately 60% of anticancer compounds and 75% of drugs for infectious diseases that are either natural products or natural product derivatives \(^{16,17}\). Despite this success, during the past couple of decades, research into natural products has experienced a steady global decline. The introduction of high-throughput synthesis and combinatorial chemistry with their promise of a seemingly inexhaustible supply of compound libraries has greatly contributed to this declining interest in the screening of natural products by the pharmaceutical industry.

Examples of important drugs obtained from plants are digoxin from *Digitalis* spp., quinine and quinidine from *Cinchona* spp., vincristine and vinblastine from *Catharanthus roseus*, atropine from *Atropa belladonna* and morphine and codeine from *Papaver somniferum*. It is estimated that 60% of anti-tumour and anti-infectious drugs already on the market or under clinical trial are of natural origin \(^{18}\). The vast majority of these cannot yet be synthesized economically and are still obtained from wild or cultivated plants. Natural compounds can be lead compounds, allowing the design and rational planning of new drugs, biomimetic synthesis development and the discovery of new therapeutic properties not yet attributed to known compounds \(^{19}\). In addition, compounds such as muscarine, physostigmine, cannabinoids, yohimbine, forskolin,
colchicines and phorbol esters, all obtained from plants, are important tools used in pharmacological, physiological and biochemical studies. In recent years, there has been growing interest in alternative therapies and the therapeutic use of natural products, especially those derived from plants. This interest in drugs of plant origin is due to several reasons, namely, conventional medicine can be inefficient effects and ineffective therapy, abusive and/or incorrect use of synthetic drugs results in side effects and other problems, a large percentage of the world’s population does not have access to conventional pharmacological treatment, and folk medicine and ecological awareness suggest that “natural” products are harmless. However, the use of these substances is not always authorised by legal authorities dealing with efficacy and safety procedures, and many published papers point to the lack of quality in the production, trade and prescription of phytomedicinal products. It is estimated that, in 1997, the world market for over-the-counter phytomedicinal products was US $10 billion, with an annual growth of 6.5%. The WHO considers phytotherapy in its health programs and suggests basic procedures for the validation of drugs from plant origin in developing countries. Eastern countries, such as China and India, have a well-established herbal medicines industry and Latin American countries have been investing in research programmes in medicinal plants and the standardisation and regulation of phytomedicinal products, following the example of European countries, such as France and Germany. In Germany, 50% of phytomedicinal products are sold on medical prescription, the cost being refunded by health insurance. In North America, where phytomedicinal products are sold as “health foods”, consumers and professionals have struggled to change this by gathering information about the efficacy and safety of these products, and new guidelines
for their registration are now part of FDA policy. In 1997, the North American market for products of plant origin reached US $ 2 billion. Thus, the modern social context and economic view of health services, the needs of the pharmaceutical market and the recognition that research on medicinal plants used in folk medicine represents a suitable approach for the development of new drugs have led to an increase in the number of publications in this field, and private and governmental institutions are now financially supporting research programmes worldwide. The NCI (National Cancer Institute, USA) has tested more than 50,000 plant samples for anti-HIV activity and 33,000 samples for anti-tumour activity. In 1993, the International Programme of Co-operation for Biodiversity (IPCB) was launched in order to promote natural products in Latin America and Africa, linking universities, industries and governments in a multidisciplinary programme for the sustained development and preservation of the environment. Large pharmaceutical companies, such as Merck, CIBA, Glaxo, Boehringer and Syntex, now have specific departments dedicated to the study of new drugs from natural sources. However, the potential use of higher plants as a source of new drugs is still poorly explored. Of the estimated 250,000-500,000 plant species, only a small percentage has been investigated phytochemically and even a smaller percentage has been properly studied in terms of their pharmacological properties; in most cases, only pharmacological screening or preliminary studies have been carried out. It is estimated that 5000 species have been studied for medical use. Between the years 1957 and 1981, the NCI screened around 20,000 plant species from Latin America and Asia for anti-tumour activity, but even these were not screened for other pharmacological activities.
Research into, and development of, therapeutic materials from plant origin is a hard and expensive task. Each new drug requires an investment of around US $100-360 million and a minimum of 10 years of work, with only 1 in 10,000 tested compounds being considered promising and only 1 in 4 of these being approved as a new drug. Upto 1992, the NCI had only found 3 plant extracts active against HIV out of 50,000 tested, and only 3 out of 33,000 plant extracts tested were found to have anti-tumour activity. Quantitative considerations regarding the average yield of active compounds and the amount of starting crude plant material required for the discovery, development and launch of a new drug on the market were presented by McChesney (1995): 50 kg of raw material are necessary to provide 500 mg of pure compound for bioassays, toxicology, and "in vivo" evaluation; full pre-clinical and clinical studies can require 2 kg of pure compounds obtained from 200 ton of raw material. The process is multi-disciplinary.

The basic sciences involved are botany, chemistry and pharmacology, including toxicology. Any research into pharmacological active natural compounds depends on the integration of these sciences. The way they are integrated and the extent of integration depend on the objectives of the study. In any case, a particular discipline should not be seen as secondary to another; quite the opposite, as each step must be carried out considering the theoretical and technical background of each of the sciences involved, otherwise the results may not be robust enough and may lead to breakdown of the process. Other fields of knowledge may also be involved if the long path from plant to medicine is taken into account. Anthropology, agronomy, biotechnology and organic chemistry can play very important roles. In addition, pharmaceutical technology is fundamental to the development of any drug, including drugs of plant origin.
Concerning drugs of plant origin, it is important to bear in mind certain conceptual distinctions. Plants can be used as therapeutic resources in several ways. They can be used as herbal teas or other home made remedies, when they are considered as medicinal plants. They can be used as crude extracts or “standard enriched fractions” in pharmaceutical preparations, such as tinctures, fluid extracts, powder, pills and capsules, when they are considered as phytopharmaceutical preparations or herbal medicines. Finally, plants can be subjected to successive extraction and purification procedures to isolate the compounds of interest, which can themselves be active and used directly as a drug, examples being quinine, digoxin and ergotamine, or they can be used as precursors (e.g. diosgenin) in hemisynthetic processes or as models for total synthesis, with well-defined pharmacological activity or structure-activity relationship studies determining a prototype drug (e.g. morphine). According to the OPS 44 a medicinal plant is (1) any plant used in order to relieve, prevent or cure a disease or to alter physiological and pathological process, or (2) any plant employed as a source of drugs or their precursors. A phytopharmaceutical preparation or herbal medicine is any manufactured medicine obtained exclusively from plants (aerial and non-aerial parts, juices, resins and oil), either in the crude state or as a pharmaceutical formulation. A medicine is a product prepared according to legal and technical procedures that is used for the diagnosis, prevention and treatment of disease and has been scientifically characterised in terms of its efficacy, safety and quality (WHO, 1992). A drug is a pharmacologically active compound, which is a component of a medicine, irrespective of its natural, biotechnological or synthetic origin.
The approach for drug development from plant resources depends on the aim. Different strategies will result in a herbal medicine or in an isolated active compound. However, apart from this consideration, the selection of a suitable plant for a pharmacological study is a very important and decisive step. There are several ways in which this can be done, including traditional use, chemical content, toxicity, randomised selection or a combination of several criteria. The most common strategy is careful observation of the use of natural resources in folk medicine in different cultures; this is known as ethnobotany or ethnopharmacology. Information on how the plant is used by an ethnic group is extremely important. The preparation procedure may give an indication of the best extraction method. The formulation used will provide information about pharmacological activity, oral versus non-oral intake and the doses to be tested. However, certain considerations must be taken into account when the ethnopharmacological approach of plant selection is chosen. For instance, each ethnic group has its own concepts of health or illness, as well as different healthcare systems. The signs and symptoms should be translated, interpreted and related to western biomedical concepts, thus allowing a focused study of a particular therapeutic property. Selection based on chemical composition uses phylogenetic or chemotaxonomic information in the search, mainly in certain genera and families, for compounds from a defined chemical class with known pharmacological activity. The search for highly specific potent drugs for therapeutic use and, more precisely, as an investigation tool in biological research has been quite productive in toxic plants. A number of important compounds now used in research came from toxic plants and several examples have been mentioned earlier. Observation of the plant’s environment has led to the isolation of active compounds,
mainly anti-bacteria and anti-insect drugs. Another method of selecting a plant is that the investigator decides on a well-defined pharmacological activity and performs a randomised search, resulting in active species to be considered for further study. The search for antitumour drugs is a good example of the use of this strategy. Finally, it is possible, and often desirable and inevitable, to use a combination of several criteria. Furthermore, apart from the chosen strategy, searching databanks and the scientific literature is crucial in finding active and/or toxic compounds that have already been identified, and can also be used as a criterion for choosing plants, e.g. if the purpose is to find a new source. However, the choice of a biological material to be screened for active compounds and the subsequent development of a drug must take into account that the exploration of natural resources should meet global and regional needs for new efficient and safe drugs, while preserving natural diversity and the environment. The present situation of exploitation of the world's vegetation may lead to the extinction of some species, which means not only the loss of interesting chemical compounds as potential drugs, but also the loss of genes, which could be of use in plant improvement or in the biosynthesis of new compounds. It is, therefore, crucial; both for the development of areas with rich flora, such as Asia and Latin America, and for the pharmaceutical industry, to protect and promote the rational exploitation of biodiversity as a source of chemical compounds that have direct biological activity or can be used for the rational planning of new drugs. By following this principle, a new understanding of sustained development emerges, involving preservation of the environment while searching for new drugs, especially in developing countries which, by coincidence, have the largest natural resources on the planet. Sensible use of these resources must be based on the
amounts available, ease of access, the possibility of preservation and replanting and the establishment of priorities in relation to a desirable pharmacological activity. If possible, consideration should be given to the use of cultivated plants, which allows the production of homogeneous material, thus guaranteeing chemical homogeneity, and the use of plants from genetic enhancement projects, which preserve species threatened with extinction 56.

The search for drugs active against tumours, viruses and cardiovascular and tropical diseases is a priority. The largest research fields, as defined by the number of publications describing bioactive plant-derived compounds in the last few years, are anti-tumour drugs, antibiotics, drugs active against tropical diseases, contraceptive drugs, anti-inflammatory drugs, immunomodulators, kidney protectors and drugs for psychiatric use 57. Taxol is both an example of the importance of natural products and of the complexity and necessity of finding alternative routes by which it can be obtained. It is the most important natural product-derived diterpene with anti-tumour activity found in recent years. Taxol is isolated from Taxus (T. brevifolia and T. bacata). However, the biggest obstacle to its clinical use is obtaining the material. In order to produce 2.5 kg of taxol, 27,000 tons of T. brevifolia bark is required and 12,000 trees must be cut down. Due to the high demand, this species of Taxus will soon be extinct if no alternative source of taxol can be developed. An economically possible and technically realistic alternative is its partial synthesis, in considerable yield, from an analogue found in other species of Taxus, as well as the production of other hemi-synthetic analogues 58,59.

Once the plant is chosen, the next step is its collection and botanical identification, and then it should be submitted to a stabilisation process. It is important that plant recollection involves a professional botanist who is able to correctly identify
the species and prepare part of the material for herbarium preservation in order to have a reference material ("voucher specimen"). Preferably, the place and date of recollection should be recorded and the information retained for further collection, if necessary. Stabilisation is usually by drying the material at ambient temperature in a shady place, but can also be carried out in an oven with controlled airflow and temperature. When the stability of the compounds is unknown or if they are known to be unstable, the fresh plant should undergo a stabilisation process consisting of freezing, lyophilisation, use of alcohol vapour, etc. The dried or stabilised plant material should then be powdered and subjected to a suitable extraction process. When the chemical nature of the compounds involved is known (once again, chemotaxonomic information and databank consultation are crucial), extraction methods should be directed at obtaining these compounds in as high a yield and purity as possible. When the chemical composition is unknown, the extraction procedure can be based on how the plant is used in folk medicine, or several extractions with solvents of increasing polarity can be performed. To obtain isolated active compounds, the plant extracts are first qualitatively analysed by thin layer chromatography (TLC) and/or other chromatographic methods and screened to determine the biological activity or to obtain a general evaluation of biological activities. For purification and isolation, the active plant extracts are sequentially fractionated, each fraction and/or pure compound being subjected to bioassay and toxicity evaluation in animals (Scheme-1).
This strategy is called bioactivity-guided fractionation. Bioassays can be performed using microorganisms, molluscs, insects, cellular systems (enzymes, receptors, etc), cell culture (animal and human), and isolated organs or in vivo (mammals, amphibians, birds, etc)\textsuperscript{63,64}. All these methods have advantages and disadvantages and the appropriate method must be carefully selected at each step of any biological study aimed at the development of a drug or the understanding of the biological basis of a particular pathology or even the discovery of the mechanism of action of already known drugs. In general, a plant extract contains low concentrations of active compounds and a large number of promising compounds, requiring the use of sensitive bioassays suitable for the wide chemical variety and small amounts of the tested samples. Tests must be simple, reproducible, fast and cheap\textsuperscript{65,66}. Furthermore, new techniques that can fulfill different needs and be adjusted to the classical pharmacological study of natural
compounds should be sought. There is also a need for the improvement and establishment of experimental models not yet extensively used in the evaluation of natural products.

After verifying the purity of an isolated active compound, the structure is determined by spectroscopic methods (UV, IR, MS or NMR) \(^6^7\). Once the chemical structure is defined, total or partial synthesis and preparation of derivatives and/or analogues can be considered, and modulation of the biological activity and definition of the structure–activity relationship can be carried out. After completing all these steps, large-scale isolation (it may necessary to collect the plant again) or partial or total synthesis is required for pharmacological evaluation in pre-clinical, clinical and toxicological trials aimed at future therapeutic use \(^6^8,^6^9\). As mentioned above, the final result of this strategy, the drug is expensive. However, the study of medicinal plants also allows their use "in natura" and/or in pharmaceutical formulations obtained from them, called *phytomedicines* or *herbal remedies*. This approach also requires efficacy and toxicity studies, but these are less time-consuming, as the steps of fractionation, purification and bioassay are basically not required or are far less complex (Scheme-2) \(^7^0\).

The Traditional Medicine Division of the WHO recognises that the centuries-old use of certain plants as therapeutic resources should be taken into account as proof of their efficacy \(^7^1\). However, the total acceptance of plant-derived drugs and phytotherapy in scientific medicine and western health systems can only occur if these products fulfill the same criteria of efficacy, safety and quality control as synthetic products \(^7^2,^7^3\). Moreover, knowledge of the main pharmacologically active plant compounds is an essential requirement for the standardisation and analysis of formulations. In the last decade, considerable effort, e.g. the Ibero-American Programme, CYTED, ESCOP (European
Scientific Cooperative of Phytotherapy) and Commission E (an independent committee on herbal remedies of German Federal Institute for Drugs and Medical Devices), has been made in trying to obtain clinical proof of efficacy, to standardise procedures for obtaining herbal remedies and to define chemical composition in order to replace crude products with modern pharmacological formulations. However, there is a long way to go!

Lack of knowledge of chemical composition, geographical distribution and environmental impact on chemical biodiversity and plant variability makes it difficult to obtain a consistent quality. Furthermore, knowledge of the effect of production methods and adjuvant compounds on the pharmacological properties of products derived from medicinal plants is still a huge research field. On the other hand, bioactivity-guided fractionation, essential when trying to isolate an active substance, may exclude plants or compounds with relevant pharmacological activities. This can occur when the effect is not caused by a single compound, but by a combination, as a result of pharmacodynamic synergism or pharmacokinetic influences. A good example of this is *Panax ginseng* in which the whole plant or its saponin fractions are more active than the isolated compounds. In addition, when only one activity is considered in pharmacological screens, it is not possible to detect other potentially useful activities. *Catharanthus roseus* was initially studied for its anti-diabetic activity described in folk medicine, but it also contains a powerful anti-tumour compound, currently in clinical use. *Ginkgo biloba* has been used for centuries in Chinese medicine to treat asthma and cough. The clinical efficacy of *Ginkgo biloba* extract was, for many years, attributed to its phenolic compounds (flavonoids and biflavonoids). The first pharmaceutical
formulations of *Ginkgo* extracts were marketed in 1960, but only a few years ago, it was found that the "standardised extract" inhibits platelet aggregation factor (PAF)-induced platelet aggregation. The compounds responsible for this effect were later isolated and identified as gingkolides A, B, C and M. Interestingly, these compounds were already known, their isolation having been described in 1932 and their chemical structure...
determined in 1967, but they were considered not to have any activity. The low yield of material, the physico-chemical characteristics of the final compound and subsequent problems, such as solubilisation of extracts and fractions in solvents compatible with the animal system, are difficulties which must be resolved in the pharmacological evaluation of these problems, in fact, can invalidate the entire study because of false negative results, interference from compounds with unspecific or cytotoxic activity, poor absorption through natural biological barriers and poor bioavailability of the products.

The limitation on the amount of material that can be obtained has been gradually overcome by the use of modern extraction, purification and isolation methods, and the development of highly specific sensitive bioassays. Auxiliary substances, such as alcohol, NaHCO₃, carboxymethyl cellulose, citric acid, DMSO, propylene glycol, polyethylene glycol and preparation of salt derivatives, are currently used to dissolve extracted materials and isolate compounds. There is an urgent need for the development and improvement of technologies for the extraction and preparation of “enriched fractions” of suitable solubility in biological fluids. In summary, research into medicinal plants and the search for plant-derived drugs require a multidisciplinary approach with integrated projects, financial and technical support, and a very carefully planned strategy. The aims should consider demands in terms of public health, preservation of Biodiversity and the technical qualification of each laboratory or research group involved. Finally, advances in technology and knowledge of natural products must be viewed not merely from the perspective of drug development, but also as a special tool for the understanding of biological phenomenon in order to contribute to the well-being of humanity.
As already discussed, phytomedicines are freely marketed and, in underdeveloped or developing countries, the use of medicinal plants is widely accepted. This can result in toxic accidents resulting from the use of plants as food or for therapy or from accidental ingestion by children or animals. Toxicity can result from highly concentrated doses or from the state of conservation of plants and the form of use. Among the various types of registered cases, it is possible to point out accidents due to mistakes of botanical identification: The use of a wrongly identified plant is common, as is the substitution of different plants for the same indication.

Plants that interfere with conventional pharmacological therapy

1. Plants containing coumarinic derivatives: These compounds can lead to haemorrhagical accidents because of their chronic use or synergistic effects with oral anticoagulants, such as dicoumarol and the sodium coumarins. Among the coumarin-rich plants widely used in folk medicine as herbal medicines and to enhance flavour are Mykania spp. ("guaco"), Melilotus officinalis ("trevo-doceamarelo") and Dypterix odorata ("fava-tonka").

2. Plants with a high tyramine content: Tyramine is a phenylethylamine found in yeast products, such as cheese and wine, which can be responsible for hypertensive accidents in patients treated with monoamine oxidase inhibitors. Mushrooms and higher plants, such as Portulacca spp. ("onze-horas"), Phoradendron spp. and Psittacanthus spp. ("erva-de-passarinho"), are also potentially dangerous.

3. Plants containing oestrogenic compounds: Ginseng (Panax spp.), used worldwide as a panacea, can have important oestrogenic effects and its use in combination with
steroid drugs is not recommended. This also applies to plants such as “inhame” (Dioscorea spp.).

4. **Plants that cause irritation and allergic problems**: Allergic reactions caused by contact with plants via pollen, secretions or volatile substances are not uncommon. The folk literature reports many plants that cause irritation; these include all species from families such as Urticaceae (*Urtica urens*), Euphorbiaceae (*Croton* spp., *Jatropha* spp., *Cnidoscolus* spp.) and Leguminoseae (*Mucuna pruriens*). Sesquiterpene lactones, found in Asteraceae, cause irritation, and plants otherwise considered harmless, such as camomille (*Matricaria recutita*) and “arnica” (*Arnica montana*), can cause dermatitis. Allergic reactions, caused by the roots of *Pfaffia* spp., are seen in workers in the herbal medicines industries, which use this plant as a substitute for *Panax* spp.

Pharmaceutical discovery is a numbers game. Thousands of chemicals must be evaluated to find a hit. The interesting agents that are identified as natural products derive from the phenomenon of biodiversity, i.e., the richness in variety of organisms in the ecosphere. A consequence of the interaction of this rich variety of organisms with each other and their environment is the evolution of diverse complex natural chemicals in the organisms that enhance their survival and competitiveness. There are literally millions of natural chemical structure types resulting from nature’s combinational chemistry effort supplying almost unimaginable chemical diversity, which yields stereochemically complex structures with diverse functional groups, molecules ideal for interacting specifically with biological target molecules. Importantly, nature has been “doing” combinational chemistry for eons, not just a decade or two, and has been selecting products from that combinational library that have specific biological advantage. Natural
products researchers have not aggressively promoted natural product preparations in terms of nature’s remarkable combinational chemistry numbers game. As Aristotle said, “Nature does nothing without purpose or uselessly.”

Natural product preparations have historically been the major source of pharmaceutical agents. Analysis of FDA new-drug approvals from 1981 to 2002 reveals that natural products continued to play a pivotal role during that time, even if the industry had turned to other discovery strategies. Indeed, more than 90% of current therapeutic classes derive from a natural product prototype and interestingly, even today, roughly two-thirds to three quarters of the world’s population relies upon medicinal plants for its primary pharmaceutical care. Those “medicinal plants” are either preparations of or...
natural product substances from plants that have potential utility as pharmaceutical agents. Historically also there were several problems associated with natural products (especially plant-derived products) that contributed to declining interest in their development within the pharmaceutical industry. Some years ago, there were significant difficulties with sourcing authenticated plant materials. It was easy to collect plants and demonstrate that their extracts had interesting biological potential. However, when researchers returned to confirm the potential and ultimately to carry out the development and commercialization of the product, failure often resulted because of inadequate documentation and loss of the original plant collections. There were also problems associated with the measurement of biological activities of natural product preparations, which ordinarily are complex mixtures of materials. Interactions among the components of the mixtures, either the antagonism by one material of another’s activity or the addition or even synergy of activities, often gave very misleading results. Purification and identification of active constituents from complex natural product mixtures containing dozens to hundreds of different chemical substances, often of quite similar chemical and physical properties, were slow and not cost-effective. Once the active constituent was isolated and purified, its chemical structure still needed to be established. These issues are compounded in that natural products are often poor pharmaceuticals; their chemical stability may be marginal; they may have poor solubility or poor bioavailability characteristics; they may not formulate well, etc., therefore not adhering to Lipinski’s Rule of Five. All of these issues have posed serious challenges. In our judgment, the issue most responsible for limited interest in plant derived natural products for pharmaceutical discovery and development has been concern over the availability of
quantities of pure chemical substances. Quantities are required initially for generation of information to understand and assess real potential of the substance for pharmaceutical application. Ultimately, the most limiting consideration is the quantity required to meet market demand should a pharmaceutical become a successful drug in the market place. Market demand can reach a scale of hundreds to thousands of kilograms per annum. It is recognized that total synthesis will not economically provide the complex natural product to meet this market demand. We believe the problems mentioned above can be overcome.

It is generally estimated that there are approximately 300,000 species of higher plants. However, some report the number to be 250,000, others estimate the number to be as high as 500,000. The disparity in the numbers partly reflects a difference in philosophy among systematic botanists. It also reflects the more aggressive exploration of unusual environments, particularly diverse environments such as the tropical rainforests, where new species of higher plants are being encountered continually. Of the approximately 300,000 species of higher plants, about 1% or roughly 3000, has been utilized for food. Of those 3000, about 150 have been commercially cultivated. In today’s market places throughout the world, unusual fruits and vegetables are beginning to appear because there is an increasing desire on the part of the world’s populations for more ‘‘exotic’’ food stuffs. However, the vast majority of caloric intake derives from about 20 species of plants. These plants represent the basis upon which the world’s population is fed, representing a very narrow foundation supporting the world’s human population. On the other hand, approximately 10,000 of the world’s plants have documented medicinal use—considerably more than the 3000 or so that have been utilized for food. Looking specifically at the utilization of plant materials in western medicine
(the US, Western Europe, etc.), it is found that roughly 150–200 of such agents are incorporated. This is still a very small percentage of all higher plants. Thus, there are potentially many more important discoveries in the plant kingdom to be exploited for pharmaceutical application.

Herbal drugs constitute a major part of traditional system of medicine. Plants above all other agents have been used for medicine from time immemorial because they have fitted the immediate personal need, are easily accessible and inexpensive. There are approximately 1250 Indian medicinal plants which are used in formulating therapeutic preparation according to Ayurveda and other traditional system of medicine 87 therapeutic preparation.

Researchers have no doubt that Nature is still the preeminent synthetic chemist and that in plants particularly, there are almost infinite reserves of fascinating chemical constituents with actual and potential effects on the human body. As such information accumulates, it becomes possible to better understand the traditional uses of plants.

Plant products can also be useful as starting material for the semi-synthetic preparation of other drugs. The main examples are plant steroids are for the manufacture of oral contraceptives and other steroidal harmones. Diosgenin from several species of Diosorea and hecogenin from Agave spp. are the main compounds used. Although interest in natural products as a source of new biologically active compounds decreased in past few decades as synthetic chemistry programmes expanded, natural products continue to form a significant proportion of drugs in current use and those of under investigation. It has been estimated that 56% of the lead compounds for medicine in the British Natural Formulary are natural products or are derived from natural products 88. Of
the top 20 best-selling pharmaceutical products in 1990 six were derived from natural products $^{89}$.

With the development of techniques of molecular biology, there has been increase in interest in the use of naturally occurring proteins as potential therapeutic agents $^{90}$. Although the wilder expectation of the ‘therapeutic protein revolution’ have not been fulfilled, several genetically engineered natural products have had a plasmogen activator is used as thrombolytic after myocardial infarctions, erythropoietin is used to treat anaemia associated with several failure and several colony stimulating factors have been marked recently for use in cancer treatment.

Production and trade of medicinal plants in India

The consumer's response to the plant based medicines has brought global resurgence in herbal trade. Continued onslaught of killer diseases like diabetes and arthritis coupled with side effects of synthetic drugs has drifted the interest of people globally from allopathy to natural/alternative system of medicine. WHO estimates the 4 billion people use herbal drugs in one form or other $^{91}$. The herbal remedies range from medicinal teas, skin care products, anti-wrinkling agents, anti-age creams and crude tablets used in traditional medicine to standardized plant extracts, phytochemicals produced in modern pharmaceutical laboratories having state of art facilities conforming to GMP standards used in modern medicine as drugs.

International trade in medicinal plants is 60 billion US dollars per year, increasing at the rate of 7 to 15% and is projected US dollar 6 trillion by the year 2050 $^{92}$. Despite of tremendous work on synthetic drugs even 25% prescription in US contain one or more constituents derived from these plant species. The primary market outlets for medicinal
Plants are phytopharmaceuticals, health, nutrition, veterinary and functional foods. Similar is the status with aromatic plants where essential oils and oleoresins derived from them are preferred over synthetic chemicals.

India has a unique position in the world where a number of traditional systems of medicine are practiced, Ayurveda being the oldest one. In addition to Ayurveda, Siddha, Unani, homeopathy and naturopathy/Tibetan system of medicine are regularly practiced for health care. These systems are plant based. Ayurveda contributes 85% followed by homeopathy (8%), Siddha (4%) and Unani (3.5%). The medicinal plants are utilized in traditional medicine, over the counter (OTC) non-prescription items involving plant parts, extracts, galenicals and phytopharmaceuticals.

A number of medicinal plants have been subjected to detailed chemical investigations and this led to the isolation of pure bioactive molecules which have been pharmacologically evaluated. These bioactive molecules are used as therapeutic agents, starting material for the synthesis of drugs, models for synthesis of pharmacologically active compounds and new reagents for molecular biology research. At present there are about 125 clinically useful drugs of known constitution, which have been isolated from...
about 100 species of higher plants. It has been estimated that about 5000 plant species have been studied in detail as possible sources of new drugs. Production of plant based drug in bulk is one of the important criteria for pharmaceutical industry in India.

In the traditional markets of India most of the medicinal plants/crude drugs are handled by traditional crude drug dealers known as Pasaris in the north. The common channel is plant collector/trader/pharmacy. The prices range from Rs 40 to Rs 1300/g. The crude extracts are sold under the Ayurveda, Unani, Siddha and local and trade names. In view of the importance of medicinal plants in health care and as revenue generating crops Govt. of India constituted National Medicinal Plant Boards in every state to co-ordinate and channelise the work on fruitful directions in this sector. The board has prioritized some 31 medicinal plants for development in different agroclimatic zones of the country. The overall production and trade in medicinal and aromatic plants should be based on a Mission Mode Approach for obtaining concrete results. There is urgent need to organize the ISM sector to cater to export needs. Proper standardization of medicinal plants and their extracts based on detailed chemical profiles, toxicological and therapeutic properties.

**Significance of Natural Products in Organic Chemistry**

Historically all organic chemistry was the chemistry of carbon compounds. Organic chemists devoted themselves to what we now refer as secondary metabolites. Some natural products among them amino-acids, carbohydrates, lipids and nucleic acids were recognized as universal constituents of living matter and play an important and primary role in metabolic reactions. Organic chemists always have been fascinated by the great diversity of these substances and particularly those that can be isolated from plants or are produced by microorganism.
Economically organic natural products of commerce (sucrose, quinine, antibiotics) are no match in value for the tremendous empire that has resulted from organic synthesis. Natural products continue to be a unique fascination not only because of their relationship to the organisms from which they are derived, not only because plants that can photosynthesize posses synthetic power that by far surpasses that of man not only because of their potential usefulness to men in their natural form a templates for organic synthetic analogue but chiefly because each new natural type reveals something of natures molecular architecture and poses new questions of how and why these compounds are produced. The compelling curiosity is to discover what compounds nature provides, but to obtain this information, it is necessary to isolate compounds from their natural source and to determine structures.

The phytochemical studies on medicinal plants have served the dual purpose of bringing new therapeutic agents, and providing useful leads for chemotherapeutic studies directed towards the synthesis of drugs modeled on the chemical structure of natural products. Moreover, they prompted studies in the correlation of chemical structure and physiological activity through functional variation in the constituents of plant material. Taking into account these facts the work under taken for the present doctoral is related to isolation and characterization of secondary metabolites from plants.