**Plant Science**: Plant Medicine—Exploring Nature's Healing Potential | The 2025 Kathy Dwyer Marble '63, MAT'65 Seminar

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### Review

## Medicinal plants: Traditions of yesterday and drugs of tomorrow

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#### Abstract

Plants have provided man with all his needs in terms of shelter, clothing, food, flavours and fragrances as not the least, medicines. Plants have formed the basis of sophisticated traditional medicine systems among which are Ayurvedic, Unani, Chinese amongst others. These systems of medicine have given rise to some important drugs still in use today. Among the lesser-known systems of medicines are the African and Australian, Central and South American amongst others. The search for new molecules, nowadays, has taken a slightly different route where the science of ethnobotany and ethnopharmacognosy are being used as guide to lead the chemist towards different sources and classes of compounds. It is in this context that the flora of the tropics by virtue of its diversity has a significant role to play in being able to provide new leads. Nonetheless the issue of sovereignty and property rights should also be addressed in line with the Convention for Biological Diversity (CBD).

This paper highlights the above, provides an overview of the classes of molecules present in plants and gives some examples of the types of molecules and secondary metabolites that have led to the development of these pharmacologically active extracts. The paper also presents some data on the use of plant products in the development of functional foods, addresses the needs for validation of plant extracts and always stressing on safety, efficacy and quality of phyto-medications.

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Keywords: Medicinal plants; Pharmacognosy; Ethnobotany; Traditional medicine; Validation; Drug discovery

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#### 1. Introduction

Throughout the ages, humans have relied on Nature for their basic needs for the production of food-stuffs, shelters, clothing, means of transportation, fertilizers, flavours and fragrances, and, not the least, medicines. Plants have formed the basis of sophisticated traditional medicine systems that have been in existence for thousands of years and continue to provide mankind with new remedies. Although some of the therapeutic properties attributed to plants have proven to be erroneous, medicinal plant therapy is based on the empirical findings of hundreds and thousands of years. The first records, written on clay tablets in cuneiform, are from Mesopotamia and date from about 2600 BC; among the substances that were used were oils of *Cedrus* species (Cedar) and *Cupressus sempervirens* (Cypress), *Glycyrrhiza glabra* (Licorice), *Commiphora* species (Myrrh) and *Papaver somniferum* (Poppy juice), all of which are still in use today for the treatment of ailments ranging from coughs and colds to parasitic infections and inflammation. Egyptian medicines report on the use of bishop's

weeds (*Ammi majus*) to treat vitiligo, a skin condition characterized by a loss of pigments. More recently, a drug ( $\beta$ -methoxypsoralen) has been produced from this plant to treat psoriasis and other skin disorders, as well as T-cell lymphoma.

The interest in Nature as a source of potential chemotherapeutic agents continues. Natural products and their derivatives represent more than 50% of all the drugs in clinical use in the world. Higher plants contribute no less than 25% of the total. During the last 40 years, at least a dozen potent drugs have been derived from flowering plants including *Dioscorea* spp.—derived diosgenin from which all anovulatory contraceptive agents have been derived; reserpine and other anti-hypertensive and tranquilizing alkaloids from *Rauwolfia* species; pilocarpine to treat glaucoma and 'dry mouth', derived from a group of South American trees (*Pilocarpus* spp.) in the Citrus family; two powerful anti-cancer agents from the Rosy Periwinkle (*Catharanthus roseus*); laxative agents from *Cassia* sp. and as a cardiotonic agent to treat heart failure from *Digitalis* species.

Approximately half (125,000) of the world's flowering plant species live in the tropical forests. Tropical rain forests continue to support a vast reservoir of potential drug species. They continue to provide natural product chemists with invaluable compounds of starting points for the development of new drugs. The potential for finding more compounds is enormous as at date only about 1% of tropical species have been studied for their pharmaceutical potential. This proportion is even lower for species confined to the tropical rain forests. To date about 50 drugs have come from tropical plants. The existence of undiscovered pharmaceuticals for modern medicine has often been cited as one of the most important reasons for protecting tropical forests, so the high annual extinction rate is a matter for concern, to say the least.

Although discovered through serendipitous laboratory observation, three of the major sources of anti-cancer drugs on the market or completing clinical trials were derived from North American plants used medicinally by Native Americans: the Papaw (*Asimina* spp.; the Western Yew Tree (*Taxus brevifolia*), effective against ovarian cancer and the Mayapple (*Podophyllum peltatum*) used to combat leukaemia, lymphoma lung and testicular cancer.

#### 2. Use of herb since antiquity to date

The vast majority of people on this planet still rely on their traditional *materia medica* (medicinal plants and other materials) for their everyday health care needs. It is also a fact that one quarter of all medical prescriptions are formulations based on substances derived from plants or plant-derived synthetic analogs, and according to the WHO, 80% of the world's population—primarily those of developing countries—rely on plant-derived medicines for their healthcare.

It is likely that the profound knowledge of herbal remedies in traditional cultures developed through trial and error over many centuries, and that the most important cures were carefully passed on verbally from one generation to another. The history of pharmacy was for centuries identical with the history of pharmacognosy, or the study of *materia medica*, which were obtained from natural sources—mostly plants but minerals, animals, and fungi.

Pharmacognosy was for the first time defined as a pharmaceutical discipline in 1815 by Seidler. The definition was "With the name Pharmacognosy we mean the science which has the task to learn everything about drugs originating from plants or animals in all aspects, except under the physiological effect, to describe them correctly and under a general vision connect this knowledge". During the 19th century, it was by far the most important pharmaceutical discipline and the mother of all present-day pharmaceutical disciplines.

Modern allopathic medicine has its roots in ancient medicine, and it is likely that many important new remedies will be discovered and commercialized in the future, as it has been till now, by following the leads provided by traditional knowledge and experiences. While European traditions are particularly well known and have had a strong influence on modern pharmacognosy in the West, almost all societies have well-established customs, some of which have hardly been studied at all. The study of these traditions not only provides an insight into how the field has developed but it is also a fascinating example of our ability to develop a diversity of cultural practices.

In some countries, the use of medicinal plants is often associated with witchcraft and superstition, because people did not have the scientific insight to explain and predict the curative action of plants. One example of such an irrational concept is the Doctrine of Signatures, elements of which are found in many of the healing cultures of the world. It is based on the assumption that the appearance of plants may give clues to their medicinal properties—it is interpreted as God's signature on the plant. Red juice and sap, for example, is associated with blood and menstrual ailments, yellow flowers with bile and jaundice, the human shape of certain roots with the female form of fertility and so on. Sometimes this concept worked: *Chelidonium majus*, contains yellow flowers and a yellow alkaloid containing latex, and has been used successfully to treat jaundice.

People who use traditional remedies may not understand the scientific rationale behind their medicines, but they know from personal experience that some medicinal plants can be highly effective if used at therapeutic doses. Since we have a better understanding today of how the body functions, we are thus in a better position to understand the healing powers of plants and their potential for their potential as multi-functional chemical entities for treating complicated health conditions. Medicinal plants typically contain mixtures of different chemical compounds that may act individually, additively or in synergy to improve health. A single plant may, for example, contain bitter substances that stimulate digestion, anti-inflammatory compounds that reduce swellings and pain, phenolic compounds that can act as an antioxidant and venotonics, anti-bacterial and anti-fungal tannins that act as natural antibiotics, diuretic substances that enhance the elimination of waste products and toxins and alkaloids that enhance mood and give a sense of well-being.

Modern allopathic usually aims to develop a patentable single compound or a 'magic bullet' to treat specific conditions. Traditional medicine often aims to restore balance by using chemically complex plants, or by mixing together several different

plants in order to maximize a synergistic effect or to improve the likelihood of an interaction with a relevant molecular target.

In most societies today, allopathic and traditional systems of medicine occur side by side in a complimentary way. The former treats serious acute conditions while the latter is used for chronic illnesses, to reduce symptoms and improve the quality of life in a cost-effective way.

#### 2.1. African traditional medicine

African traditional medicine is the oldest and perhaps the most diverse of all medicine systems. Africa is considered to be the cradle or Mankind with a rich biological and cultural diversity marked regional difference in healing practices. Unfortunately, the systems of medicines are poorly recorded and remain so to date. Yet the documentation of medicinal uses of African plants is becoming increasingly urgent because of the rapid loss of the natural habitats of some of these plants because of anthropogenic activities. The African continent is reported to have one of the highest rates of deforestation in the world. The paradox is that it is also a continent with a high rate of endemism with the Republic of Madagascar topping the list at 82%.

African traditional medicines in its varied forms, is a holistic involving both the body and the mind. The healer typically diagnoses and treats the psychological basis of an illness before prescribing medicines to treat the symptoms. Famous African medicinal plants include *Acacia senegal* (Gum Arabic), *Agathosma betulina* (Buchu), *Aloe ferox* (Cape Aloes), *Aloe vera* (North African Origin), *Artemisia afra* (African wormwood), *Aspalanthus linearis* (Rooibos tea), *Boswellia sacra* (Frankincense), *Catha edulis* (Khat), *Commiphora myrrha* (Myrrh), *Harpagophytum procumbens* (Devil's Claw), *Hibiscus sabdariffa* (Hibiscus, Roselle), *Hypoxis hemerocallidea* (African potato), *Prunus africana* (African Cherry). Madagascar by herself has contributed with the *Catharanthus roseus* (Rosy Periwinkle) and has the potential of contributing more in view of the diversity of her flora and fauna.

#### 2.2. American traditional medicine (North, Central and South)

#### 2.2.1. North America

In the US, just like in any other cultures, the indigenous healer or Shaman approaches illnesses by addressing both the physical and spiritual dimension of diseases. These Shamanistic ceremonies involve chanting, dancing and other rituals aimed at expelling evil forces so that the patient or the community as a whole can be healed. Early settlers learnt from native practices and they eventually adopted many of the herbal remedies, which later formed the basis of the Pharmacopeia of the United States. Among the famous medicinal plants of the United States are the Echinacea (*Echinacea purpurea*) and Goldenseal (*Hydrastis canadensis*). During most of the 20th century, herbs or botanicals have been regarded with skepticism and the practice of herbal medicine went into decline. Plants were viewed mainly as a potential source of pure chemical compounds for the development of medicine.

In recent years, herbs and botanicals have become very popular in the US and Canada but they are still considered as nutritional supplements rather than medicines in their own rights.

#### 2.2.2. Central and South America

Just like Africa, Central and South American countries have a rich and diverse healing cultures and which are poorly known and have not been properly recorded. They will no doubt be a source of new herbal remedies in the years to come. South and Central America have made enormous contributions to agriculture and a large number of food crops (maize, potatoes, tomatoes, pumpkins, cassave, peanuts, sweet potato) originate from there. Traditional Indian medicinal herbs are also used extensively but the influence of Spanish, European, Indian and African is obvious. Famous examples of medicinal plants are: *Cinchona pubescens* (Peruvian bark), *Erythroxylum coca* (Coca), *Ilex paraguariensis* (Maté), *Myroxylon balsamum* (Tolu balsam), *Paullinia cupana* (Guarana), *Peumus boldus* (Boldo), *Psidium guajava* (Guava), *Spilanthes acmella* (Brazilian cress), *Tabebuia impetiginosa* (Lapacho) and *Uncarina tomentosa* (Cat's claw).

#### 2.3. Australian and Southeast Asian medicine

This region has witnessed a resurgence of interest in traditional medicine and many countries now promote research into medicinal plants as a potential source of new remedies. The Aborigines had a complex healing system but much of the traditional knowledge in Australia was lost before it could be systematically recorded. In contrast, many healing places like Malaysia, Thailand, Vietnam, New Zealand, Borneo, and the Polynesian Islands remain intact and are being recorded and developed. A strong Chinese influence is being observed in most countries. Among the well-known medicinal products originating from this region are *Croton tiglium* (Purging croton), *Duboisia hopwoodii* (Pituri), *Eucalyptus globulus* (Bluegum), *Melaleuca alternifolia* (Tea tree), *Myristica fragrans* (Nutmeg and Mace), *Piper methysticum* (Kava kava), *Strychnos nux-vomica* (Strychnine), *Styrax benzoin* (Benzoin) and *Syzygium aromaticum* (Cloves).

#### 2.4. Ayurvedic medicine (Indian Traditional Medicine)

Ayurveda is perhaps, the most ancient of all medicinal traditions is probably older that the traditional Chinese medicine. It is considered to be the origin of systemized medicine. It is actually a practical and holistic set of guidelines to maintain balance and harmony in the system. Dioscorides (who influenced Hippocrates) is thought to have taken many of his ideas from India. Ancient Hindu writings on medicine contain no references to foreign medicines whereas Greek and Middle Eastern texts do refer to ideas and drugs of Indian origin.

Ayurveda is derived from the Indian words 'Ayar' (Life) and 'veda' (Knowledge or Science) and hence means the Science of Life. Following the system would help

ensure a long life, which is considered to be the instrument for achieving righteousness (*dharma*), wealth (*artha*) and happiness (*sukha*).

In India, knowledge and wisdom have been passed on from one generation to the next through songs and poems, which scholars and physicians had to learn by heart and recite. The Veda is an ancient text in four parts (Rig Veda, Sama Veda, Yajur Veda and Atharva Veda), the earliest of which date back to 2000 years BC. The principles of Ayurvedic medicine and the medicinal plants uses of herbs are contained in thousands of poetic hymns in the Rig Veda.

The first school to teach Ayurvedic medicine was at the University of Banaras in 500 BC and the great Samhita (or encyclopedia of medicine) was written. 700 Years later, another great enclyclopedia was written and these two together form the basis of the Ayurveda.

Ayurveda is similar to Galenical Medicine in that it is based on bodily humours (*dosas*) and the inner life force (*prana*) that is believed to maintain digestion and mental activity. The living and the non-living environment, including humans, is composed of the elements earth (prithvi), water (jada), fire (tejac), air (vaju) and space (akasa). For an understanding of these traditions, the concept of impurity and cleansing is also essential. Illness is the consequence of imbalance between the various elements and it is the goal of the treatment to restore his balance.

Famous Ayurvedic medicinal plants include *Azadirachta indica* (Neem), *Centella asiatica* (Gotu Kola), *Cinnamomum camphora* (Camphor), *Elettaria cardamomum (ela* or cardamomum), *Rauwolfia serpentina* (Indian snake root), *Santalum album* (Sandalwood), *Terminalia* species (Myrobolan) and *Withania somnifera* (Aswargandha).

#### 2.5. Chinese traditional medicine

The civilizations of China and India were flourishing when only modestly sophisticated cultures were developing in Europe. Expectedly writings on medicinal plants and the aesthetics of vegetation were numerous. This ancient system of medicine, believed to be more than 5000 years old, is based on two separate theories about the natural laws that govern good health and longevity, namely *yin* and *yang*, and the five elements (*wu xing*).

The legendary emperor Shen Nung discussed medicinal herbs in his works which were probably written 2500 years B.P. instead of their traditional date of 3500 B.P. By the Traditional Chinese medicine was systematized and written between 100 and 200 BC. The most complete reference to Chinese herbal prescription is the Modern Day Encyclopedia of Chinese *materia medica* published in 1977. It lists nearly 6000 drugs out of which 4800 are of plant origin.

Yin and Yang denotes opposites that complement each other. The five-element theory is similar to the four humours and elements of the Greeks or the three humours of Ayurveda. The five elements are earth, metal, water, wood and fire each of which is linked to the main organ systems of the body (respectively the spleen, lungs, kidney, liver and heart), the emotions (reflection, grief, fear, anger, joy), the climates (damp, dry, cold, windy, cold), the seasons (late summer, autumn, spring, summer) and tastes (sweet, pungent, salty, sour, bitter) and so on. Medicine is used to restore or maintain balance between these elements and to grant vital energy (qi) which has both yin and yang aspects. Treatment is therefore based not only on symptoms but also on pattern of imbalances, often detected by taking the pulse or observing the patient's tongue. Warming or hot herbs such as ginger, and cinnamon, are used to treat ailments associated with cold symptoms such as cold hands, abdominal pains and indigestion.

In common with Western and African traditional medicines, Chinese herbs are usually given in fixed mixtures or formulas of up to 20 herbs, carefully prepared according to traditional recipes contained in ancient compendia. There are hundreds such recipes being used alongside with Western Medicines. As in other healing cultures, traditional recipes are used preferentially against chronic illnesses while acute or serious illnesses are cured by Western Medicines.

The spread of traditional Chinese medicine to most continents has undoubtedly contributed to the current popularity of herbal medicines throughout the world. Examples of famous Chinese medicinal herbs are *Angelica polymorpha* var. *sinensis* (dang gui), *Artemisia annua* (qing hao), *Ephedra sinica* (ma huang), *Paeonia lactiflora* (bai shao yao), *Panax ginseng* (ren shen) and *Rheum palmatum* (da huang).

#### 2.6. European medicine

In the ancient Western world, the Greeks contributed significantly to the rational development of the use of herbal drugs. However, the European healing system is said to have originated with Hippocrates (460–377 BC) and Aristotle (384–322 BC), whose own ideas were rooted in ancient beliefs from India and Egypt. The philosopher and natural scientist, Theophratus (~300 BC), in his *History of Plants*, dealt with the medicinal qualities or herbs, and noted the ability to change their characteristics through cultivation. Dioscorides, a Greek physician (100 AD) during his travels with Roman armies, recorded the collection, storage and the use or medicinal herbs and Galen (130–200 AD) who practiced and taught pharmacy and medicine in Rome, published no less than 30 books on these subjects, and is well known for his complex prescriptions and formulas used in compounding drugs, sometimes containing dozens or ingredients ("galenicals").

Greek and Roman medicine was based on the belief that the world is composed of four elements—earth, wind, fire and water. Each of these has its corresponding humours, linked to the four vital fluids in the body. The four humours—blood, phlegm, black bile and yellow bile, influence both health and temperament (respectively sanguine, phlegmatic, melancholic and choleric). In order to restore balance, drastic measures such as bloodletting (reduce excess blood) and purging (to remove excess black bile) was used. The four humours were also associated with cold, heat, dampness and dryness and each of these had a corresponding range of cold, hot, damp or dry herbs that were supposedly able to restore imbalances. European tradition also had many regional influences on local folk practices and traditions.

One of the most powerful influences was the famous book *De Materia Medica*, written by the Greek physician Dioscorides in the first century AD. It is generally accepted to be the first European herbal and was the standard reference in Europe for more than 1000 years, providing the base for most of the later herbals. As early

as AD 800, medicinal plants were cultivated according to a standardized layout in monasteries in Central Europe. One of the famous healers of this era was Hildegard of Bingen (1098–1179). In later years a Swiss alchemist known as Paracelsus (1493–1541) emphasized the importance of the correct dose for medical treatments.

Herbal medicine is part of everyday life in many countries in Europe and to this day has remained popular as a sophisticated and rational method of treating ailments, often considered to be supportive than curative. In several European countries to date, the use herbal tea is still very popular. In addition to these, 'natural products' taken in their crude form (unprocessed) as teas or decoction, sophisticated phytomedicines (standardized and formulated extracts of plants, often subject to rigorous testing in humans) remain a popular alternative to medicinal products derived from pure synthetic chemicals.

A large number of traditional herbal remedies in Europe have become widely known as a result of commercialization and a number of active compounds have been isolated from medicinal plants and are used today as single chemical entities.

#### 2.7. Classical Arabic and North African traditional medicine

The oldest written information in the Arabic traditions comes from the Sumerians and Akkadians of Mesopotamia, thus originating from the same areas as the archeological records of Shanidar IV (Heinrich et al., 2004).

#### **Documents of Shanidar IV**

The earliest documented record, which presumably relates to medicinal plants, dates from 60,000 BCE in the grave of the Neanderthal man from Shanidar IV, an archeological site in Iraq. Pollen of several species of plants, presumably used as medicines, was discovered among which are: *Centaurea solstitialis* (Asteraceae), *Ephedra altissima* (Ephedraceae), *Althea* sp. (Malvaceae) amongst others.

Although this may not be a finding with no direct bearing on the culture of Shanidar, these species or closely related ones from the same genus, are still important today in the phytotherapy of Iraq and also known from other cultural traditions. These species may well be typical for the Neanderthal people and may also be part of a tradition for which Shaidar IV represents the first available record.

The Middle East is known as the cradle of civilisation and many plants grown nowadays have been domesticated in this region. The Babylonians, Assyrians and Sumerians recorded herbal remedies in cuneiform writing on numerous clay tablets. Of special interest is the Code of Hammurabi (ca. 1700 BC), a comprehensive set of civil laws carved in stone and commissioned by the King of Babylon. It lists several medicinal herbs. Similar documents have survived several millennia in Egypt. The Egyptians documented their knowledge (including medical and pharmaceutical) in wall-paintings of tombs dating from the Old Kingdom and on papyrus, the latter being made from *Cyperus aquaticus*. The most important of these writings is the Ebers Papyrus, which originates from around 1500 BC is reported to contain ancient medicinal knowledge from before 3000 BC. This famous 20 m Papyrus scroll reputedly found in a tomb is inscribed in Egyptian hieroglyphics and named after Prof. Ebers Georges at Thebes in 1872. It was brought in 1873 and deposited at the University of Leipzig and two years later G. Ebers published a facsimile edition. The Ebers Papyrus is a medical handbook covering all sorts of illnesses and includes empirical as well as symbolic forms of treatment. The diagnostic precision documented in this text is impressive.

If during the Dark and Middle Ages (5–12th Centuries, AD), the monasteries in countries such as England, Ireland, and Germany have preserved the remains or this Western Knowledge, it was the Arabs who were responsible for the preservation or much or the Greco-Roman expertise, and for expanding it to include the use or their own resources, together with the Chinese and Indian herbs, till then unknown to the Greco-Roman world. The Arabs were the first to establish privately owned drug stores in the 8th century, and the Persian pharmacists, physician, philosopher and poet, Avicenna, contributed much to the sciences or pharmacy and medicine throughout the works such as *Canon medicinae*, regarded as the "final codification of all Greco-Roman medicine".

*Canon medicinae* include elements of other healing cultures and forms the basis for a distinct Islamic healing system known today as *Unani-Tibb*.

Among the famous medicinal plants of the Middle East and Egypt are: Allium cepa (Onion), Astracantha gummifera (Tragacanth), Carthamus tinctorius (Safflower), Carum carvi (Caraway), Ferula assafoetida (Asofoetida), Lawsonia inermis (Henna), Papaver somniferum (Opium poppy), Peganum harmala (Syrian rue), Prunus dulcis (Almond), Punica granatum (Pomegranate), Rosa x damascena (Damask Rose), Ricinus communis (Castor Oil Plant), Salvadora persica (Toothbrush tree), Senna alexandrina (Senna), Sesamum indicum (Sesame), Trachyspermum ammi (Ajowan), Trigonella foenum-graecum (Fenugreek) and Vitis vinifera (Grape).

#### 3. Ethnobotany and medicine

#### 3.1. Ethnobotany

The term '*Ethnobotany*' was first used by Harshberger in 1896. He defined it as a study of 'plants used by primitive and aboriginal people'. The term was broadened by Robbins, Harrington and Freire-Marreco, in 1916 and they suggested that the science of ethnobotany should include the investigation and evaluation of the knowledge of all phases of life amongst primitive societies and of the effects of the vegetal environment upon the life customs, beliefs and history of these tribal peoples. Twenty-five years later, Jones (1941) advanced a more concise definition: "The study

of the interrelationships of primitive men and plants". Schultes in 1967, expanded this to include 'the relationships between man and his ambient vegetation'.

#### 3.2. The search of new drugs through ethnomedicine

As mentioned above, plants have formed the basis for traditional medicine systems, which have been used for thousand or years in countries such as China (Chang and But, 1986) and India (Kapoor, 1990). The use of plants in traditional medicine systems of many cultures has been extensively documented. These plant-based systems continue to play an essential role in health care and the World Health Organisation estimates that 80% or the world's inhabitants continue to rely mainly on traditional medicines systems of their health care. Plant products also play an important role in health care systems of the remaining 20% of the population, mainly residing in developed countries. Analysis of the data on prescriptions dispensed from community pharmacies in the US from 1959 to 1980, indicates that 25% contained plant extracts or active principles derived from higher plants and at least 119 chemical substances, derived from 90 plant species, can be considered as important drugs currently in use in one or more countries (Farnsworth et al., 1985).

Of these 119 drugs, 74% were discovered as a result of chemical studies directed at the isolation of the active substances from plants used in traditional medicine. In addition, the use of so-called complementary or alternative herbal products has expanded in recent decades.

The isolation of the anti-malarial drug, quinine from the bark of *Cinchona* species (e.g. *C. officinalis*) was reported in 1820 by the French pharmacists, Caventou and Pelletier. The bark had long been used by indigenous groups in the Amazon region for the treatment of fevers, and was first introduced into Europe in the early 1600s for the treatment of malaria.

There are four basic ways in which plants that are used by tribal peoples are valuable for modern medicine:

- 1. Plants from the tropics are sometimes used as sources of direct therapeutic agents (e.g. the alkaloid D-tubocurarine is extracted from the South American jungle liana *Chondrodendron tomentosum* is widely used as a muscle relaxant in surgery. Chemists are still unable to produce this drug synthetically in a form that is has all the attributes of the natural product and therefore collection from the wild is still relied upon. Surprisingly harvesting of medicinal plants is often less costly than artificial drug synthesis. Another good example to illustrate this feature is Reserpine, an important hypotensive agent extracted from *Rauwolfia*. The synthesis of this molecule would cost three times as much as opposed to collection).
- 2. Tropical plants are also used as sources of starting points for the elaboration of semi-synthetic compounds. An example of this would saponin extracts that are chemically altered to produce sapogenins necessary for the manufacture of steroidal drugs. Until relatively recently, 95% of all steroids were obtained from extracts of neo-tropical yams of the genus *Dioscorea*.

- 3. Flora from the tropics can serve as sources of substances that can be used as models for new synthetic compounds. Cocaine from Coca plants, *Erythroxylum coca*, has served as model for the synthesis of a number of local anaesthetics such as procaine. New and unusual chemical substances found in plants will continue to serve as 'blueprints' for novel synthetic substances and will prove to be increasingly important in the future.
- 4. Plants can also be used as taxanomic markers for the discovery of new compounds. From a phytochemical standpoint, the Plant Kingdom has been investigated in a haphazard manner; some families have been relatively well-studied while others have been almost completely overlooked. For example, many uses have been documented for the Liliaceae and the family is known to be rich in alkaloids. Little, on the other hand is known on the Orchidaceae. Some plants from the family have been investigated because of their close relationship to the Liliaceae. Research has shown that they are not only rich in alkaloids but that some of the alkaloids are unique and could prove useful in the future.

# 4. Value of ethnobotany and ethnopharmacology to the search for new biodynamic compounds or new leads?

Ethnobotany and Ethnopharmacology are interdisciplinary fields of research that look specifically at the empirical knowledge of indigenous peoples concerning medicinal substances, their potential health benefits and their health risks associated with such remedies. As can be seen, many of the plant-derived pharmaceuticals and phytomedicines currently in use, were used by native people around the world. Some of this knowledge has been documented and codified or studied scientifically.

Also of the hundreds of thousands of species of living plants, only a fraction has been investigated in the laboratory. This poor understanding of plants is particularly acute in the tropics. Brazil, which has some 55,000 species of plants, has reports on only 0.4% of the flora!

The importance of ethnobotanical inquiry as a cost-effective means of locating new and useful tropical plant compounds cannot be over emphasized. Most of the secondary plant compounds employed in modern medicine were 'first' discovered through ethnobotanical investigation.

Of the 119 pure chemical compounds extracted from higher plants used in medicine throughout the world and 74% of these compounds have the same or related use as the plants from which they were derived.

The Rosy Periwinkle (*Catharanthus roseus*, Apocynaceae) represents a classical example of the importance of plants used by local peoples. This herbaceous plant, native to southeastern Madagascar, is the source of over 75 alkaloids, two of which are used to treat childhood leukaemia and Hodgkin's disease with a very high rate of success. This species was first investigated in the laboratory, mainly because of its use by local people as an oral hypoglycaemic agent. Like the *Catharanthus*, many drugs that are commonly used today (e.g. aspirin, ephedrine, ergometrine, tubocurarine,

digoxin, reserpine, atropine etc.) came through the use of indigenous medicine that is through the bioscientific investigations of plants used by people throughout the world. Table 1 lists just a few of the many examples of drugs derived from plants.

Thus it can be seen that the investigation of plants used for medicinal purposes by 'unsophisticated' peoples can provide us with new biodynamic compounds that may have important applications in our society.

When one considers the importance of medicinal plants in the developing countries, it is not surprising that most of the world's populations depends on traditional medicine for their primary health care needs. In many cases, developing countries simply cannot afford to spend millions of dollars on imported medicines. Several African and Asian nations are now encouraging traditional medicines as an integral component of their public health care programs. Indigenous medicines are relatively inexpensive and locally available and are readily accepted by the local population.

#### 5. Secondary plant metabolites in drug discovery

Although natural products, particularly secondary metabolites, have formed the basis of medicines and the presence of these compounds in the biochemistry of the plant is very often difficult to justify. It has been suggested that these compounds may have been synthesized by the plant as part of the defense system of the plant, e.g. plants are known to produce phytoalexins as a response to attack by bacteria and fungi. The presence of highly toxic natural products has also been highlighted in some animals namely the Amazonian frogs so as to deter predation by other animals. Whatever the reasons for the presence of these compounds in Nature, they provide an invaluable resource that have been used to find new drug molecules. Fig. 1 gives an indication of the development of new drugs from leads coming from natural products.

#### 6. Methods used in natural product chemistry

Spectroscopic methods coupled with good extraction techniques like chromatography, have contributed to the phenomenal success of natural product chemistry over the past 50 years. A sound isolation strategy has helped in the isolation and characterisation of many bioactive molecules. Nowadays, bioassay-guided fractionation of medicinal plants is a feature of routine in the attempt to isolate bioactive components from natural sources. These techniques are not only being restricted to plant sources but they are also being applied to microbial and even fungal sources of metabolites.

In practice as soon as the material is collected, in the case of plants, it needs to be identified by a taxonomist so as to ascertain the correct identity of the material. Voucher specimens for herbarium specimens are kept. Various parts of the plant are collected separately (leaves, flowers, stem, wood, bark, root, root bark etc.) and are dried quickly in drying cabinets. Quick drying avoids degradation of the components

Botanical drugs used in traditional medicine and which have given useful modern drugs								
Botanical names	English names	Indigenous use	Origin	Uses in biomedicine	Biologically active compounds			
Adhatoda vasica	_	Antispasmodic, antiseptic, insecticide, fish poison	India, Sri Lanka	Antispasmodic, oxytocic, cough suppressant	Vasicin (lead molecule for Bromhexin and Ambroxol)			
Catharanthus roseus	Periwinkle	Diabetes, fever	Madagascar	Cancer chemotherapy	Vincristine, Vinblastine			
Condrodendron tomentosum	_	Arrow poison	Brazil, Peru	Muscular relaxation	D-Tubocurarine			
Gingko biloba	Gingko	Asthma, anthelmintic (fruit)	Eastern China	Dementia, cerebral deficiencies	Ginkgolides			
Harpagophytum procumbens	Devil's claw	Fever, inflammatory conditions	Southern Africa	Pain, rheumatism	Harpagoside, Caffeic acid			
Piper methysticum	Kava	Ritual stimulant, tonic	Polynesia	Anxiolytic, mild stimulant	Kava pyrones			
Podophyllum peltatum	May apple	Laxative, skin infections	North America	Cancer chemotherapy, warts	Podophyllotoxin and lignans			
Prunus africana	African plum	Laxative, 'Old man's disease'	Tropical Africa	Prostate hyperplasia	Sitosterol			

 Table 1

 Botanical drugs used in traditional medicine and which have given useful modern drugs



Fig. 1. Medicinal agents made from Nature's lead molecules.

by air or by microbes. Sometimes the plant samples are lyophylised at high vacuum, but again care must be exercised so as to avoid the excessive loss of volatile components as the latter may also exhibit interesting biological activities.

Once the material has been dried to constant weight, it is ground up to smaller particles and extracted usually using a gradient solvent extraction. Nonetheless, in the process or validation or the ethnobotanical information, the utilisation by the lay people must be mimicked as far as possible so that the same natural bioactive products are extracted.

This is a very critical stage in the work as if the extraction technique is not adapted, it may result in the loss of access to the active components. Additionally, improper extraction methods may result in the degradation of the natural product.

Numerous extraction techniques are available namely:

*Cold extraction*: whereby the plant material is extracted in solvent of differing polarity at room temperature. It allows for maximum extraction of most components.

*Hot percolation*: the plant is heated in the solvent usually under reflux. This extraction method allows for the extraction of a large number of metabolites, from the most insoluble material like the waxes to the lipophilic natural products.

Supercritical fluid extraction: In this extraction technique, some gases behave as though they are liquids with solvating properties, under pressure. In this instance, the gas commonly used is carbon dioxide, which has the advantage of being able to be blown away from the extractant after the extraction is over. The polarity of carbon dioxide can be enhanced by adding a modifying agent such as carbon tetrachloride.

*Soxhlet extraction*: Perhaps the most widely and commonly used extraction technique for the extraction of natural product. The polarity gradient of the solvent is applied. Although some components may be destroyed in the process, it is still the best method or extraction used in natural product chemistry. Once the extraction is complete, the extractant is usually concentrated under vacuum, for large volumes or solvents and 'blown down' under nitrogen for small volumes, ensuring at the same time that volatiles are not lost. Aqueous extracts are generally freeze-dried and stored at -20 °C as this low temperature reduces the degradation of the bioactive natural product. Extraction protocols may sometimes be modified depending on the type of molecules being extracted, e.g. sometimes acids may be added to extract alkaloids as their salts.

#### 6.1. Isolation methods

Once the extract has been obtained, the activity within can be demonstrated by bioassay methods using both the crude extract or by using the fractionated extracts. Fractionation has the added advantage of getting to the biologically active material faster. One of the simplest separation methods is partitioning which is a widely used method as an initial extract purification step. A combination of solvents—miscible and immiscible ones are used to separate the phytochemicals making up the extract. This method relies on the ability or the components to be either soluble in water or in the organic phase.

#### Chromatography

Chromatograhic techniques have been instrumental in the separation of natural products. Some of the techniques are discussed here. One of the fastest and most widely used chromatographic techniques is Thin Layer Chromatography (TLC).

TLC: This method employs glass or aluminium plates pre-coated with the sorbent (e.g. Silica gel) to varying thickness depending on the amount to be loaded. The compound mixture is loaded both in the preparative or analytical plates at around 2 cm from the bottom and lowered in a tank containing the solvent. The latter migrates up the plates and separates the compound mixture according to the polarity of the components. Several reagents are available for visualization of the separated materials.

TLC has the advantage of being a highly cost-effective qualitative technique in as much as a large number or samples can be analysed or separated simultaneously. The few drawbacks include poor detection and control or elution compared to High Performance Liquid Chromatography (HPLC).

*HPLC*: This method is very popular and widely used for the analysis and isolation of bioactive natural products. The analytical sensitivity is further enhanced depending on the detector that is being used. The detectors can be UV detection such as a photodiode array (PDA), which enables the acquisition of UV spectra of eluting peaks between 190 nm to 800 nm. PDA UV detection has the advantage of detecting even compounds with poor UV characteristics and this is particularly useful in the analysis of natural products such as terpenoids or polyketides, which may not necessarily have chromophores that will rise to a characteristic UV signature.

Coupled with electronic library searching of compounds along with the 'fingerprinting' of biologically active extracts, HPLC becomes a very powerful quality control technique of herbal medicines. It has now become a tool of choice for the analysis of a majority of natural products in the pharmaceutical industry. It suffers from one drawback in the sense that it is expensive both from the machine and consumable view-points.

Another 'clean up' chromatographic technique that has become increasingly popular and useful is Gel Chromatography or sometimes referred to as Size Exclusion Chromatography (SEC). This technique employs cross-linked dextran which upon contact with a suitable solvent swells up to form a gel matrix. The latter contains pores of a finite size allowing smaller molecules to be retained and excluding larger molecules.

This method is excellent for separating out fatty acids, chlorophyll etc prior to biological assays. This is a non-destructive method for the recovery of a high quantity or extract. It is a method or choice for large molecules such as proteins, polypeptides, carbohydrates etc.

#### 7. Assays used in evaluating the activity of extracts

#### 7.1. Bioassay

#### 7.1.1. Requirements for screening medicinal plant material

Bioassay is a very crucial stage in assessing the pharmacological actions of plant extracts and their ethnomedical uses. In the initial stages, in vitro testing have priority over in vivo studies involving laboratory animal models. This decision is usually based on scientific, economic and ethical grounds. In vivo studies may be preferable at later stages of the research project but still depends on the amount and the nature of evidence or bioactivity already collected by means or, in vitro, studies and the quest for additional information under life conditions. Bioactive components that are candidates for therapeutic application will still have to undergo extensive clinical and toxicological screening programmes before they can be registered as medicines.

#### 7.1.2. Common pharmacological screening methods

There are many types of pharmacological screens. They are specific for bacteria, fungi, protozoa, intestinal worms, viruses etc. The efficacy of compounds against health problems such as cancer and inflammation is often probed while the effect on physiological and anatomical systems such as reproduction, digestion etc can be judged. Among the commonly used assays are the brine shrimp, antimicrobial (bacteria and fungi) screens amongst others:

• Brine shrimps are small aquatic animals that can be grown in solutions similar to seawater. In order to test the potential toxicity of the plant—and thus its probability of containing anti-cancer agents—measured amounts of plant extracts are added to containers holding known numbers of brine shrimps. The surviving brine shrimps are counted after 6 and 24 h, and the acute and chronic LD<sub>50</sub> values are calculated, respectively; this corresponds to the concentration or the compound in solution that kills 50% of the brine shrimps.

• Bacteria are grown on agar medium in Petri dishes. When measured amounts of a plant extract are placed on paper disks or in wells set on the surface of the bacteria-inoculated agar under sterile conditions, after 18–24 h, bacteria-free circles can be seen around the disk or wells, indicating that the extract has inhibited the microbes. Plant extracts can also be tested for phototoxic and fungicidal activity against yeasts and other strains of fungi.

It is worthpointing out that advances in plant drug research will provide information on bioactivity in terms of molecular interactions with other target groups. Developments in the fields of genetics, molecular biology, bioinformatics and techniques used in determining the steric structure of plant metabolites and target macromolecules appear important now. Fundamental understanding of molecular biodiversity seems important in the process of using plant resources for drug development. Animal models are being replaced by testing on cell cultures and this implies that smaller amounts of test compounds will be needed. Also they will provide information at cellular level. This can only lead to more extensive studies on medicinal plants.

After the screening process, a series of active extracts will need to be dereplicated (i.e. ascertain that no compounds are present that are already known and are present in the extract assayed). Dereplication ensures that novelty is brought into the isolation process and it thus ensures that new compounds are identified and will eventually get patented.

#### 8. Characterization and structure elucidation of bioactive compounds

Once the biological evaluation has been performed and the separation of the natural product has been achieved, the chemist will try to attempt the elucidation of the compound. Structure elucidation depends on classical spectroscopic techniques such as: Nuclear Magnetic Resonance (NMR) 1-D and 2-D Proton NMR as well as C-13 NMR, Infra Red (IR), Mass Spectrometry (MS) and X-Ray analysis.

For exploring nature's chemodiversity, the situation has changed dramatically in recent years by the introduction of high throughput screening (HTS) methods. By using molecular targets, a large number of samples (up to 100,000 in 24 h) can be screened for a single activity. Obviously synthetic chemists are not able to produce such numbers or new compounds. Their answer was the development of combinatorial chemistry and testing mixtures of compounds obtained through this novel solid-phase chemical synthetic methods.

HTS offers new possibilities for natural products. It allows rapid screening of large number of extracts and it is very suitable for bioassay-guided fractionation, which in the past was the major bottleneck in studies of active compounds in plant extracts.

Powerful chromatographic methods in combination with HTS are now a very efficient way to new leads for drug development. Searching data bases such as Chemical Abstracts, NAPRALERT as well as the Dictionary of Natural Products ensure that no time is wasted on re-investigating existing and known molecules. These databases lead to primary literature information ranging from molecular weights to spectroscopy data, which help to recognize common metabolites at an early stage.

Structure-elucidation is crucial in assessing the biological activity of the molecule as it is a well-known fact that biological activity depends to a large extent on the 3-D arrangement of functional groups on the molecule.

#### 9. Important plant families having given molecules/drugs of importance

*Apiaceae* (previously known as the Umbelliferae): Originating from the temperate regions of the world, this family, comprising of some 3000 mostly herbaceous species, has given rise to many of the common spices and herbs used to date. The plants are aromatic and rich in essential oils.

Among the common herbs used medicinally are the following:

- Caraway (*Carum carvi*)—Used against bloats
- Coriander (Coriander sativum)—Carminative
- Fennel (Foeniculum vulgare)-Mildly Carminative
- Anis (Pimpinella anisum)-Expectorant, Spasmolytic, Carminative

*Apocynaceae*: This family has wide distribution both in temperate and tropical regions of the world. Among the world famous species is the Rosy Periwinkle (*Catharanthus roseus*). Members of this family are well known for their alkaloidal contents with potent pharmacological activity.

Araliaceae: This family is of over 700 species widely distributed in both the tropical and subtropical parts of the world. The best-known European specimen is the Ivy (*Hedera helix*). Another world famous species of this family is the Ginseng (*Panax ginseng*). In the tropics there are several endemic species of the Araliaceae family, which are classified as being heterophyllous plants—plants having differing foliage shapes and sizes on the same plant. The reasons for this phenomenon are still unknown. Phyto-pharmacologically, these plants are characterized by the presence of saponins, triterpenoids and some acetylenic compounds. *Panax ginseng* owes its pharmacological activity to the triterpenoids (ginsengosides) and the secretolytic activity effect of *H. helix* is partly responsible to the presence of saponins.

Arecaceae (Palmae): The Palm family which comprises of some 2700 species almost exclusively woody is an important family as it includes many species widely used as food and over the past years at least one of its members has become medicinally important. The Sawpalmetto (*Serenoa repens*) is now being used for difficulty in micturition in benign prostrate hyperplasia in the early stages. Phytopharmacologically, the plant is known to accumulate polyphenols, some relatively simple alkaloids (especially pyridine derivatives) and steroidal saponins as well as fatty acids. The pharmaceutical use of the Sawpalmetto seems to be due to the presence of a relatively large amount of the triterpenoid— $\beta$ -sitosterol. Asphodelaceae: (Members from this family have often been grouped under the Liliaceae family). This family, with about 600 species, is widely distributed in South Africa and some species occur in the Mediterranean regions. The best-known members in this family are *Aloe vera* and *A. ferox*. The genus Aloe is characterized by the presence of polysaccharides accumulating in the leaves as well as anthranoids and anthraglycosides (aloe-emodin). Contrary to other related families, the Asphodelaceae do not accumulate steroidal saponins.

Asteraceae: (The 'Daisy' family and previously known as the Compositae family). This large family comprises of some 25,000 species and 1400 genera and is distributed and is well represented in most ecosystems except for Antarctica. Phytochemically, this family is characterized by the presence of polyfructanes (especially inulin) as storage carbohydrates as opposed to polysaccharides, in the perennial taxa. In some taxa, some segments of the family accumulate sesquiterpene lactones (e.g Parthenolides), which are important natural products responsible for the pharmacological activity of many botanical drugs e.g. Fever few (*Chrysanthemum parthenium*) and Arnica (*Arnica montana*). Some taxa accumulate pyrrolizidine alkaloids e.g. in the Senecio species and these compounds are known for their hepatotoxic effects. Other taxa accumulate unusual diterpenoids e.g. the diterpene glycoside—Stevioside, known for its intensely sweet taste.

#### **Gymnosperms**

Ginkgoaceae: (The Ginkgo family)



Gingko biloba

This family is perhaps the most ancient of the seed bearing plants and had been widely distributed in the Mesozoic. Only one species exists today. Phytochemically, this plant is characterized by the presence of the gingkolides which are unusual two-ringed diterpenoids with three lactone functions. Biflavones and glycosylated flavonoids are other groups of typical natural products.



Ginkgolide C

Hypericaceae



This family of some 900 species, distributed both in the temperate and tropical regions, has gained importance by virtue of at least one of its members—St. John's Wort (*Hypericum perforatum*). This plant has become one of the most important medicinal plants in the 20th century. Phytochemically, several members of this family contain resins, balsam and the flowers contain naphthodianthrones e.g. hypericin and pseudohypericin—characteristic of the *Hypericum* genus.



*Lamiaceae*: This family, with over 5000 species, has been one of the most important ones in the contribution of medicinally important and culinary species. They are aromatic and have also yielded commercially important essential oils. Several species accumulate Rosmarinic acid and other derivatives of Caffeic acid. Rosmarinic acid is one some pharmaceutical importance because of its non-specific complement activation and inhibition of leukotrienes (leading to an anti-inflammatory effect).



Rosmarinic acid

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*Papaveraceae*: This rather small family has produced a multitude of pharmacologically important genera (e.g. *Chelidonium*, *Glaucium*, *Papaver*). This family is particularly rich in the isoquinoline alkaloids including morphine, papaverine, codeine, thebaine and noscapine. Other alkaloids present include the benzylisoquinoline (papaverine, noscapine).

Some of the isolated molecules from these plants have been particularly useful and are:

- Celidonine (Chelidonium majus)-employed as a cholagogue.
- Morphine and other alkaloids (*Papaver somniferum*)—well known analgesic and potent narcotics.



Morphine

*Piperaceae*:



The Pepper family consists of herbs and shrubs comprise some 2000 species is mostly restricted in the tropics. An important genus is the *Piper*. This family is also characterized by pungent acidic amides such as piperine and also essential oils present in many members.

Some of the isolated molecules from these plants are:

• Kavain (*Piper methysticum*)—well known in Oceania for conditions of nervous anxiety.

*Rhamnaceae*: This family consists of trees and comprises some 900 species. An important genus of this family is the *Rhamnus*. This family is known to accumulate anthraquinones. Alkaloids of the benzylisoquinoline type and the cyclo-peptide type are also known from many taxa.

• Rhamnus species (R. purshiana and R. frangula) are used as strong purgatives.

*Rubiaceae*: This large family of over 10,000 species, has yielded one of the most important stimulants—Coffee (*Coffea arabica* and *C. canephora*). Another medicinally important species having been brought into the Old World is the Cinchona bark, extracted from *Cinchona* sp. The Rubiaceae is characterized by the presence of iridoids (a group of monoterpenoids), alkaloids (including indole alkaloids such as Quinine from *Cinchona* species), methylxanthines such as caffeine, theobromine, theophylline and anthranoids.

*Rutaceae*: This family comprises of some 1700 species distributed throughout the world but the tropics are particularly rich in them. Perhaps the most well known examples from this family are the Citrus with orange, lime, grapefruit, mandarin etc.

This family is characterized by essential oils found in the secretory cavities in the pericarp and parenchyma. Alkaloids are also found and among them are the benzyltetrahydroxyisoquinoline, acridone and imidazole types. Acridone alkaloids have so far only been reported from the Rutaceae. Furano- and pyrano-coumarins e.g. bergapten as well as other simple coumarins have been isolated from *Citrus* species.

• Pilocarpine (Pilocarpus jaborandi) has been used in ophthalmology.

Many species from this family have been used as aromatic plants in perfumery and also as foods.



Pilocarpine

*Solanaceae*: This family of 2600 species, includes some of the most important staples—the Potato (*Solanum tuberosum*) and several other medicinal and toxic plants, known for the highly active natural products. Important genera of this family include Atropa, Datura and Hyoscyamus. Some of the pharmacologically active molecules isolated from these genera include the following:

Atropine (*Atropa belladonna*) Nicotine (*Nicotiana tabaccum*) Zingiberaceae:



Curcuma longa

This family, distributed throughout the tropics, is rich in essential oils with terpenes (borneol, camphene and cineole (all oxygen-containing monoterpenes), sesquiterpenes (zingiberene) and phenyl propanoid derivatives (cinnamic acid derivatives). Important medicinal plants from this family are the following:

Ginger (*Zingiber officinalis*)—used against a large variety of illnesses including travel sickness.

Elaiti (Elettaria cardamomum)-used as a spice but also as a medicine.

Turmeric (*Curcuma longa*)—used as a spice and useful against inflammatory and liver diseases in most Asian medical systems for a large variety of illnesses.

#### 10. Plant parts used

*Root*: The fleshy or woody parts of many species are used medicinally. Roots may be fibrous (*Urtica dioica* or *U. radix* of the Urticaceae family, Stinging nettle), solid (*Glycyrrhiza glabra* of the Leguminosae family, Liquorice) or fleshy (*Harpagophytum procrumbens* of the Pedaliaceae family, Devil's claw).

*Rhizome*: The rhizome is a woody or fleshy elongated stem that usually grows horizontally below the ground, forming leaves above the ground and roots into the ground. Medicinally important rhizomes include Kava kava (*Piper methysticum* of the Piperaceae family) and the Ginger (*Zingiber officinalis* of the Zingiberaceae family).

*Bulb*: A bulb is the fleshy structure made up of numerous layers of bulb scales which are leaf bases. Bulbs popular for medicinal use include the Onion and Garlic (*Allium cepa* and *A. sativum* respectively, both of the Liliaceae family).

*Tuber*: A tuber is a swollen, fleshy structure below the ground, usually of stem origin but often partly stem and partly roots. The African Potato (*Hypoxis* sp. of the Hypoxidaceae family) is a well known example.

*Bark*: The bark is the outer most protective layer of a tree trunk and is formed by layers of living cells just above the wood itself. There are usually high concentrations of the active ingredients in the bark and several examples of the bark exists e.g. the Quinine bark (*Cinchona* sp., Rubiaceae) and Cinnamon and Camphor (*Cinnamo-mum camphora* and *C. camphora* both of the Lauraceae family).

*Wood*: The wood is the thick stem or the wood itself. Important examples of useful woods include Sandalwood (*Santalum album* of the Santalaceae family).

*Leaf*: The leaves can sometimes be used alone or mixed with the petiole. Example of plants where only the leaves are used is the Gingko *(Gingko biloba* of the Ginko-aceae family).

*Aerial parts*: All parts of the plant found above the ground are referred to as the aerial parts. Very often the plants, which have useful aerial parts, are harvested when flowering. One such example is the St. John's Wort (*Hypericum perforatum* of the Hypericaceae family).

*Flowers*: Flowers are very commonly used and popular in traditional medicine. Several flowers commonly used in medicine include the Clove (*Syzygium aromaticum*, Myrtaceae), Camomille flower (*Chamaemelum nobile*, Asteraceae), Roselle (*Hibiscus sabdiriffa*, Malvaceae), and the Marigold (*Calendula officinalis*, Asteraceae). *Fruit*: Among the most commonly used seeds, one finds the following: Anis seeds (*Pimpinella* anisum) and the Fennel fruit (*Foeniculum vulgare*) both of the Apiaceae family. In some instances, the fruit peel is used specifically, e.g. Pomegranate (*Punica granatum*, Punicaceae) and the fruit peel of Citrus fruits (Citrus sp., Rutaceae).

*Seeds*: Seeds are contained in the fruit and in some instances are used by themselves. Examples exist for the use of the seeds e.g. Castor oil (*Ricinus communis*, Euphorbiaceae), and the seeds of the Fennel (*Foeniculum vulgare*, Apiaceae).

*Gum*: Gums are solids consisting of mixtures of polysaccharides. They are watersoluble and are partially digested by humans. Gums sometimes flow from a damaged stem as a defense mechanism or sometimes as a protective system against the invasion of bacteria and fungal rots. Well known examples of gums are Gum Arabic (*Acacia Senegal*, Leguminosae), Benjoin (*Terminalia bentzoe*, Combretaceae) and Aloe gel (*Aloe vera* gum of the Liliaceae family mixed with water).

*Resins*: Resins are excreted from specialized cells or ducts in plants. They consist of a mixture of essential oils and polymerized terpenes, usually insoluble in water. Well-known examples of resins since Biblical times include Frankincense (*Boswellia sacra*) and Myrrh (*Commiphora myrrha*) both of the Burseraceae family.

*Fatty oils*: These are non-volatile, insoluble oils pressed either from the seeds or from the fruits of plants. Oils are often referred to as acylglycerides because they are derived from glycerol molecules. Olive oil is a useful example in as much as these oils over their own therapeutic potential are also used in carriers as liquid formulations and ointments.

*Essential oil:* These are volatile oils usually extracted from plants through a process of either steam distillation or microwave extraction. They consist of terpenes (mono- and sesquiterpenoids and coumarins). They are of considerable importance as active ingredients of medicinal plants. Well known examples include Peppermint oil (*Mentha x piperita*, Lamiaceae), Ylang ylang oil (*Cananga odorata*, Annonaceae) amongst others.

#### 11. Dosage forms

*Extracts* are made either in liquid, powdered or viscous forms from the crude mixtures plant parts. The chemical compounds can then be extracted from plant material using water or organic solvents such as alcohol (ethanol). As a result, the extract contains only the soluble fractions of the plant material (usually about 20% of the total weight) and the non-soluble (fibrous) residues (about 80%), which are discarded. Volatile oils are extracted by steam distillation or less often by solvent extraction. The herb to extract ratio (HER) is typically 5:1 for normal extracts, or say 100:1 for a herb with 1% essential oil. The extract draws its origins from tradition but it is still commonly used today.

*Mixtures* are products with medicinal properties and which contain 2 or more plants or herbs that can act individually, additively or even synergistically to restore

or maintain health. In Chinese, Indian and African Traditional medicines, medicinal plants are typically used in mixtures.

*Teas* or *Infusions* prepared by steeping herbs in boiling water. They are called Teas because of the similarity in preparations.

*Decoction* refers to a preparation that is made by adding cold water to the required amount of the drug and then boiled and allowed to simmer for 5-10 min. The mixture is strained afterwards.

*Maceration* refers to a preparation made by adding cold water to the required amount of the drug, which is allowed to soak at room temperature for 6–8 h before it is strained.

*Juice* is prepared by crushing the freshly harvested plant parts and then expressing the juice. The product can be pasteurized or treated at ultra-high temperatures to extend their shelf-life.

*Syrup* is a preparation containing about 66% sucrose and generally has a viscous consistency. Saturated sugar solutions are free of microorganisms because no free water for microbial growth is available. Syrups are mainly used as flavouring agent to mask an unpleasant taste of other ingredients. When used as a cough mixture, it is slowly sipped so as to allow maximum contact with the inflamed mucous membrane.

*Tincture* refers to an alcoholic solution (usually 30–70% water) prepared from medicinal plant materials. The herbal mixture is extracted for an indefinite period after which it is pressed and/or strained to separate the liquid and solid materials. A mother tincture is prepared by using 70% ethanol and the solution is then diluted with clean water to a predetermined herb to extract ratio. Glycerides may be prepared by using glycerol as opposed to the alcohol.

*Medicinal essences* are volatile compounds dissolved in alcohol or alcoholwater mixtures. Medicinal spirits are produced by mixing aromatic herbs with alcohols and then recovering the alcohol and volatile components by steam distillation.

*Capsules* are usually small but soft or hard containers normally made from gelatin. They contain medicinal products or extracts in a predetermined dose and also to protect them from air, light and moisture.

*Tablets* There are two types or tablets—uncoated and coated tablets. Uncoated ones are made by compression of powdered active material after addition of a suitable inert excipient or binder (to provide bulk) and sometimes also other additives to improve colour, flavour or disintegrators to ensure that the tablet rapidly dissolves when placed in water.

*Pills* are made by dividing semi-solid preparations into smaller portions of predetermined size or weight, or rolling the portions before allowing them to harden. The manufacture of pills is now fully mechanized.

*Suppositories* are tablet-like products usually oblong to oval in shape and that are intended for inserting into the rectum, vagina or urethra and left there to melt. Herbal products are rarely used in this form.

*Ointments* are usually semi-solid preparations aimed at external application. They usually contain medicinal substances in a suitable carrier substance (watery or oily solvents).

*Poultice* is a paste made from the crushed fresh plant. It can either be mixed with oil, alcohol or simply made in water and applied on the parts of the body.

#### 12. Modes of administration

*Oral*: A decoction, infusions, tinctures, syrups and tablets are most often taken orally and sometimes sublingually.

*Nasal (Smoking, snuffing or steaming)*: Essential oils suspended in hot liquids or powdered materials may be snuffed so that the active compounds are resorbed through the mucosa. Smoke from burning materials is inhaled and the active compounds resorbed into the lungs (in the same way that nicotine is absorbed into the lungs).

*Topical*: Lotions, oils or creams containing extracts of medicinal plants are applied directly to the skin, where the active compound is absorbed.

*Rectal*: The liquid preparations can be administered as enemas. The active compounds are absorbed by the mucuous membrane of the rectum. In some instances, depending on the nature of the extracts, it may be desirable to by-pass the stomach.

Bathing: Herbs or herbal extracts may be added to bath water.

Sub-cutaneous or intramuscular injections: Some phytomedicines (often pure chemical entities derived from medicinal plants) are injected into the bloodstream. Interestingly, sometimes some compounds are completely inactive when taken through the mouth and yet highly active when injected. One good example of the usefulness of such compounds is the Menispermaceae alkaloids used often as muscle-relaxant dart poisons. The meat killed from the killed animals is harmless to eat.

#### 13. Phytochemistry and the modes of action of plant metabolites

Phytochemistry deals with the chemistry of plant metabolites and their derivatives. The metabolic system of a plant may be regarded as being constituted of regulated processes within which biochemical conversions and mass transfer take place. Understanding in this field has advanced to a stage in which definite metabolic processes, biosynthetic pathways and their interconnections are distinguished and studied in the context of their function and genetic control.

The metabolic performance of living organisms can be distinguished into primary metabolism and secondary metabolism. Primary metabolism is associated with fundamental life processes common to all plants. It comprises processes such as photosynthesis, pentose cycle, glycolysis, the citric acid cycle, electron transport, phosphorylation and energy regulation and management. Primary metabolites are produced and converted molecular entities, needed in anabolic pathways to build, maintain and reproduce the living cell. In catabolic pathways, primary metabolites (and food products) provide the chemical energy and precursors for biosynthesis. Primary and secondary metabolisms are interconnected in the sense that the biosynthesis of accumulating secondary metabolites can be traced back to ubiquitous primary metabolites.

However, in contrast to primary metabolites, secondary metabolites represent features that can be expressed in terms of ecological, taxonomic and biochemical differentiation and diversity. The biosynthesis and accumulation of secondary metabolites provide a basis for biochemical systematics and chemosystematics. In addition, the wide molecular diversity of secondary metabolites throughout the plant kingdom represents an extremely rich biogenic resource for the discovery of novel drugs and for developing innovative drugs. Not only do plant species yield raw material for useful compounds; the molecular biology and biochemistry provide pointers for rational drug development.

Primary and secondary metabolites can be classified on the basis of their chemical structure into much the same categories of chemical compounds: Carbohydrates, Lipids, Amino acids, Peptides, Proteins, Enzymes, Purine and pyrimidine derivatives. Within such compounds classes, secondary metabolites generally show greater individuality and diversity in their molecular structure than primary metabolites. Certain compound classes also appear to be extraordinarily rich in secondary metabolites, e.g. the structurally diverse groups of alkaloids, phenolics, acetogenins and terpenoids. Ubiquitary primary metabolites belonging to these compound classes seem to be restricted to only a limited number of key compounds functioning as biosynthetic precursors.

Most of the plant compounds that have been found to be medicinally useful and interesting tend to be secondary metabolites. Also despite enormous structural diversity, Nature just uses a few building blocks, e.g. Shikimic acid and Shikimate, to create this chemo-diversity. Shikimic acid accounts for the synthesis of many aromatic amino acids including phenylalanine, tyrosine, tryptophan as well as organic acids like benzoic and gallic acids and aldehydes like vanillin, benzaldehyde.

The basic building blocks are the acetate  $(C_2)$ , isoprenoid  $(C_5)$  and phenylpropanoid  $(C_9)$  units. The acetate unit is used in the polyketide biosynthesis and is particularly well developed in microorganisms. The isoprenoid pathways lead to all terpenoids by coupling two or more  $C_5$  units. The terpenoids are found in all organisms.

The phenylpropanoid pathway is most typical for plants and it is based on phenylalanine and tyrosine via cinnamic acid and this pathway leads to amongst others lignin and lignans. In combination with three acetate units the C<sub>9</sub> unit leads to the flavonoids and the anthocyanins, well known for their role in flower colour. Quite a number of natural product groups can be constructed from the amino acid phenylalanine, in particular, flavonoids, coumarins, lignans etc. all of which possess a common substructure based on aromatic 6-C ring (C<sub>6</sub> unit) with a 3-C chain (C<sub>3</sub> unit) attached to the aromatic ring (see figure).

Many reactions can occur to this  $C_9$  unit including oxidation, reduction, methylation, cyclization, glycosylation (adding of sugar molecules) and dimerization all of which contribute to the value of natural products as a resource of biologically active molecules enhancing at the same time, to the structural complexity of the molecule with the presence of chirality and functionality.



The metabolites synthesized by the plants are of many different types and include the following:

#### Carbohydrates

These compounds are the first products plant produce by photosynthesis from water and carbon dioxide. They can be grouped into sugars, and polysaccharide. The sugars are either monosaccharides such as glucose, fructose or oligosaccharides containing 5–6 monosaccharide units. Monosaccharide can be trioses, tetroses, pentoses and heptoses and are  $C_3-C_7$  compounds. The polysaccharides are macromolecules containing a large number of monosaccharide residues.

Carbohydrates make up a large portion of plant biomass, e.g. cellulose is part of the cellular framework and starch as food reserves. Sugars can unite with a wide variety of compounds to form glycosides, increasing the water solubility of the compounds. Glycosides vary in their chemical structure and pharmacological activity due to their aglycone component.

In addition to their use as bulking agents in pharmaceuticals, carbohydrates have recently been recognized to have useful pharmacological properties. Several polysaccharides exhibit immuno-modulatory, anti-tumour, anticoagulant (e.g. heparin), hypoglycaemic or antiviral activities. The various carbohydrate products traded include fibre, cellulose and its derivatives, starch (glucose polymers) and its derivatives, dextrins, fructans (fructose polymers; e.g. Inulin), algenic acids, agar and gums.

#### Lipids

Vegetable oils are major sources of  $\beta$ -sitosterol, which is a steroid drug precursor. One vegetable oil, obtained from groundnut, yields lecithins, which are used to enhance food digestibility. Lecithins are also used in pharmaceutical formulations. Recently, some vegetable oils have been found to be rich in  $\gamma$ -linoleic acid, which is a precursor of prostaglandin, leukotrienes and thromboxanes. All these compounds are involved in platelet aggregation and inflammatory processes. Only members of Onagraceae, Saxifragaceae and Boraginaceae contain  $\gamma$ -linoleic acid (Padua et al., 1999).



γ-Linoleic acid

Vegetable oils are significant in both the food and pharmaceutical industries. Some are used as solvents for lipid-soluble drugs such as vitamins and antibiotics. Others e.g. almond and olive oils are used in cosmetics. Castor oil is also well known for its purgative activity, but has fallen out of favour because of its unpleasant taste. *Acetogenins* 

These molecules are long-chain aliphatic compounds with over 35 carbon atoms, ending with a  $\gamma$ -lactone, most often un-saturated and cyclized into one or two tetrahydrofuran rings that may or may not be adjacent. They have been isolated from members of the Annonaceae (e.g. *Annona, Goniothalamus, Rollinia* and *Uvaria*). Their potential applications are linked to their anti-tumour (e.g. asimicin, bullatacine), antibacterial (e.g. cherimolin) and insecticidal (e.g. asimicin, annonin, annonacin) properties.



Annonacin

#### Amino acids and their derivatives

The function of amino acids is not only the building blocks of proteins but they are also considered to be a form of nitrogen storage (e.g. Cannovanine, Hemoarginine) and a germination inhibitor. The few studies of the pharmacological activities of amino acids include reports of cucurbitine being used as a taenicide. Many toxic amino acids have been identified; examples include  $\beta$ -( $\gamma$ -L-glutamylamino) proprionitrile and g-N-oxalyl-L- $\alpha$ , $\beta$ -diaminopropanoic acid which are responsible for the toxicity of grass pea (*Lathyrus sativus* L.) that brings about osteolathryrism and neurolathyrism in livestock and mimosine from *Leucaena* inhibiting proteins and nucleic acid synthesis which results in livestock losing appetite and weight and their growth being inhibited (Padua et al., 1999).

#### (i) Cyanogenic glycosides

Cyanogenic glycosides are compounds also derived from amino acids. Enzymatic or acid hydrolysis of these compounds yield hydrocyanic acid, which is the toxic principle. Biosynthetically, the aglycones of cyanogenetic glycosides are derived from L-amino acids. Cyanogenic glycosides are prevalent in the families of the Rosaceae, Leguminosae, Graminae, Araceae, Euphorbiaceae and Passifloraceae. Examples include linamarin, amygdalin, prunasin.

#### (ii) Sulphur-containing compounds

The pharmacological significance of sulphur-containing compounds like allein, allicin, ajoene, and other related compounds have been demonstrated. Initially isolated from garlic, allicin and ajoene (the latter is a condensation product of allicin) exhibit many biological activities, including anti-hypercholesterolaemic, anti-platelet aggregation, anti-hypertensive, fibrinolytic and anti-fungal activities. Recently diallyl cysteine, an odourless active ingredient of garlic was found to be biosynthesized. (iii) *Lectins* 

Lectins are proteins or glycoproteins that are able to bind with the carbohydrate moiety on cell membranes in a specific and reversible fashion, without displaying enzymatic activity. Most lectins in higher plants are located in seeds. They are commonly found in legumes such as groundnut, soya bean and common bean. Some lectins have the ability to agglutinate red blood cells of a specific blood group. These lectins are referred as phytohaemagglutinin. The haemagglutination activity is important in immunological studies. Some lectins are toxic, e.g. Ricin from Castor (*Ricinus communis* L.) seeds and abrin from Jequirity bean (*Abrus precatorius* L.) seeds.

#### (iv) Enzymes

Plant-derived enzymes used as drugs include papain (*Carica papaya*) and bromelain (*Ananas comosus*). Both are proteolytic enzymes useful as an anti-inflammatory drug. Ficin, extracted from the Fig (*Ficus carica* L.) has similar properties.

#### Alkaloids

The term 'alkaloid' has been defined as a cyclic organic compound containing nitrogen in a negative oxidation state, which has limited distribution in living organisms. Based on their structures, alkaloids are divided into several subgroups: nonheterocyclic alkaloids and heterocyclic alkaloids, which are again divided into 12 major groups according to their basic ring structure. Mescaline is an example of a non-heterocyclic or pseudo-alkaloid, Tetrandrine is another example of a bisbenzylisoquinoline alkaloid while Solasodine is a triterpene alkaloid.



Free alkaloids are soluble in organic solvents and react with acids to form watersoluble salts. There exceptions like Berberine, which is a quartenary ammonium alkaloid. Most alkaloids are solids except for Nicotine, which is a liquid.

Alkaloids, usually having a marked physiological action on humans or animals, are believed to be waste products and a nitrogen source. They are thought to play an important role in plant protection and germination and to be plant growth stimulants. Alkaloids are more common in dicotyledons than in monocotyledons. Families reported to be rich in alkaloids are: Liliaceae, Amaryllidaceae, Apocynaceae, Berberidaceae, Leguminosae, Papaveraceae, Ranunculaceae, Rubiaceae and Solanaceae.

Alkaloids are pharmaceutically significant, e.g. morphine as a narcotic analgesic, Codeine in the treatment of coughs, Colchicine in the treatment of gout, quinine as an anti-malarial, quinidine as an anti-arrythmic and L-hyoscyamine (in the form of its racemic mixture known as atropine) as antispasmodic and for pupil dilation.

Recently, the extracts of *Erythroxylum pervillei* collected in Madagascar, have yielded nine tropane alkaloids out of which seven are new ones. Six of the new compounds (pervilleins A-F) were found to reverse multidrug resistance (MDR), using a KB-VI (vinblastine-resistant oral epidermoid carcinoma). These compounds show promise as they are novel inhibitors of the MDR phenotype (Kinghorn et al., 2003).

#### Phenols and phenolic glycosides

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Phenols are among the largest group of secondary metabolites. They range from simple structures with one aromatic ring to complex polymers such as tannins and lignins. Example of phenolic classes of pharmaceutical interests are:

(1) Simple phenolic compounds: These compounds have a monocyclic aromatic ring with an alcoholic, aldehydic or carboxylic group. They may have a short hydrocarbon chain. Capsaicin, isolated from *Capsicum* sp., is a vanillyl amide of isodecenoic acid and is marketed as an analgesic. Eugenol is widely used in dentistry due to its anti-bacterial and anti-inflammatory and local anaesthetic activities.

(2) *Tannins*: The chemistry of these compounds is very complex. The distinction made in the literature between hydrolysable and condensed tannins is based on whether acids or enzymes can hydrolyse the components or whether they condense the components to polymers. Although not altogether watertight, this distinction largely corresponds to group based on gallic acid and those based on flavane-related components. Several vegetable tannins have been discovered but only the tanning constituents of the most important groups will be reported here, i.e. the group of gallotannins and ellagitannins, the group of proanthocyanidins. Gallotannins and ellagitannins are esters of gallic acid or its dimers digallic acid and ellagic acid with glucose or polyols.

Proanthocyanidins are oligomers of 3-flavanols (catechins) and 3,4-flavandiols (leucoanthocyanidins).

These phenolic compounds are biosynthesized via the Shikimic acid or acetate pathway.



Tannins are able to react with proteins. Upon being treated with a tannin, a hide absorbs the stain and is protected against putrefaction and thereby becomes converted into leather. Although tannins are widespread in plants, their role is still unclear. They may be an effective defence against herbivores. Tannins are used against diarrhoea and as an antidote in poisoning by heavy metals. Their use declined after the discovery of hepatotoxic effects of absorbed tannic acids. Recent studies have reported that tannins have anti-cancer and anti-HIV activities.

(3) Coumarins and their glycosides: Coumarins are, shikimate-derived, benzo- $\alpha$ -pyrone derivatives that are present in plants both in a free state and as glycosides. They have limited distribution in the plant kingdom and have been used in chemo-taxonomy in order to classify plants. They give a characteristic odour of new-mown. They are found in the following plant families: Apiaceae, Rutaceae, Asteraceae and Leguminosae. Common derivatives are: Umbelliferone, Herniarin, Aesculetin, Scopoletin, Fraxin and Chicorin.

Some coumarins are phytoalexins and are manufactured by the plant in the event of an infection by bacteria and fungi. Scopoletin is among such compounds and are synthezised by the potato plant (*Solanum tuberosum*) when attacked by fungi. Aesculin extracted from the Horse Chestnut (*Aesculus hippocastanum*) goes into the phytotherapeutic preparations for the treatment of capillary fragility.



(4) *Quinones*: Quinones are oxygen-containing compounds that are oxidized homologues of aromatic derivatives and are characterized by a 1,4-diketo-cyclohexa-2,5-diene pattern (paraquinones) or by a 1,2-diketo-cyclohexa-3,5-diene pattern (ortho-quinones). In naturally-occurring quinones, the dione is conjugated to an aromatic nucleus (benzoquinones), or to a condensed polycyclic aromatic system: naphthalene (naphthoquinones), anthracene (antraquinones), 1,2-benzanthacene (anthracyclinones), naphthodianthrene (naphthodianthrone), pyrelene, phenanthrene and abietane-quinone. Naphthoquinones and anthroquinones have some importance medicinally.

(i) *Naphthoquinones*: These are yellow or orange pigments from plants. Biosynthetically, the naphthoquinones are derived almost exclusively from the Shikimic acid pathway. Naphthoquinones are found in families like: Bignoniaceae, Ebenaceae, Droseraceae, Juglandaceae, Plumbaginaceae, Boraginaceae, Lythraceae, Proteaceae and Verbenaceae.

The pharmaceutical significance of this group is limited except for a few examples like Plumbagin isolated from *Plumbago* species, exhibit anti-bacterial and cytotoxic properties. Lawsone from Henna (*Lawsonia inermis* L.) is a powerful fungicide and a well-known hair dye.


Plumbagin

(5) *Flavonoids*: Flavonoids are compounds that are responsible for the colour or flowers, fruits and sometimes leaves. The name refers to the Latin word '*flavus*' meaning yellow. Some may contribute to the colour by acting as co-pigment. Flavonoids protect the plant from UV-damaging effects and play a role in pollination by attracting animals by their colours.

The basic structure of flavonoids is 2-phenyl chromane or an Ar–C3–Ar skeleton. Biosynthetically they are derived from a combination of the Shikimic acid and the acetate pathways. Small differences in basic substitution patterns give rise to several subgroups. In the plant, flavonoids can either occur as aglycones or as O- or C-glycosides.



Basic structures of some flavonoids

Recently, flavonoids have attracted interest due to the discovery of their pharmacological activities as anti-inflammatory, analgesic, anti-tumour, anti-HIV, antiinfective (anti-diarrhoeal, anti-fungal), anti-hepatotoxic, anti-lipolytic, anti-oxidant, vasodilator, immunostimulant and anti-ulcerogenic. Biologically active flavonoids comprise of hesperidin and rutin for decreasing capillary fragility and quercetin for its anti-diarrhoeal activity.

(i) Anthocyanins: Anthocyanins are the compounds responsible for bright colours of most flowers and fruits. These water-soluble pigments occur as glycosides (anthocyanins *sensu stricto*) and their aglycones (anthocyanidins). They are derived from the 2-phenyl benzopyrylium cation, more commonly referred to as the flavylium cation. Cyanidin is an example of an anthocyanin.



Cyanidin (Cyanidin-3,5-diglucoside)

The application of anthocyanins is as food additives, e.g. in beverages, jams and confectionary products. The pharmacological activities are similar to flavonoids; for instance for decreasing capillary permeability and fragility, and as anti-oedema.

*Phloroglucinols* are derivatives of 1,3,5,-trihydroxybenzene which are found in e.g. Cannabis sativa L. This compound is a well-known stimulant of the Central Nervous system. Tetrahydrocannabinol and its derivatives influence behaviour inducing euphoria and relaxation at low doses. Sometimes hallucination and tinitus are observed. Other effects are bronchodilatation and a lowering of intra-ocular pressure.

(6) *Lignans and related compounds*: Lignans and related compounds are derived from condensation of phenyl propane units. Neolignans are also condensation products of phenylpropanoid units. Norlignans are probably specific to gymnosperms and have a C17 skeleton.

Lignins are substances deposited at the end of the formation of the primary and secondary cell walls. Chemically, they are polymers arising from the copolymerisation of alcohol with a *p*-hydroxycinnamic structure (*p*-hydroxycinnamyl, coniferyl or sinapyl alcohol). Lignins are always combined with polysaccharides.

The pharmacological activity of lignans is anti-tumour. Kadsurenone, a neolignan, exhibits anti-allergic and anti-rheumatic activity. The major application of Lignin is as a precursor of vanillin, which is widely used in the food industry.

(7) *Terpenoids and steroids*: Terpenoids and steroids are derived from the isoprene (a 5-C unit), which is biosynthesized from acetate via mevalonic acid.

#### Monoterpenes

Monoterpenes are the most simple of constituents in the terpene series and are C10 compounds. They arise from the head to tail coupling of two isoprene units. They are commonly found in essential oils. Iridoids and pyrethrins are included in this group. Examples of monoterpenes commonly found in essential oils are found below:



Iridoids are monoterpenes characterized by a cyclopenta [C] pyranoid skeleton, also known as the iridane skeleton. Secoiridoids, which arise by cleavage or 7,8-bond or the cyclopentane ring, are also included in the iridoids.



Examples of secoiridoids are the bitter constituents or gentian e.g gentiopicroside, amarogentin and esters of sweroside and swertiamarin.

Other examples of irregular monoterpenes arising from the non-classic coupling of isopentenyl pyrophosphate and dimethylallyl pyrophosphate are the class or compounds known as the Pyrethrins. These compounds are toxic to coldblooded animals such as fish, amphibians and insects and are widely used as insecticides.

The pharmacological properties of iridoids are quite limited except for the analgesic and anti-inflammatory activities of the Harpagosides (*Harpagophytum* sp.).

#### (i) Sesquiterpenes

Sesquiterpenes are also constituents of essential oils of many plants, e.g. bisabolol, humulene and caryophyllene. Sesquiterpene lactones are well known as bitter principles. They occur in families like the Asteraceae.

These compounds possess a broad range of activities due to the  $\alpha$ -methylene- $\gamma$ -lactone moiety and epoxides. Their pharmacological activities are anti-bacterial, anti-fungal, anthelmintic, anti-malarial and molluscicidal. Examples are Santonin, which is used as an anthelmintic and as an anti-malarial.



# (ii) Diterpenes

Diterpenes constitute a vast group of  $C_{20}$  compounds arising from the metabolism of 2E,6E,10E-geranylgeranyl pyrophosphate. They are present in animals and plants. These compounds have some therapeutic applications. For example, Taxol and its derivatives are anti-cancer drugs. Other examples are Forskolin, which has anti-hypertensive activity. Zoapatanol is an abortifacient while Stevoside is a sweet-ening agent. Taxol is another very famous diterpene.

# (iii) Triterpenes and steroids

Triterpenes are  $C_{30}$  compounds arising from the cyclization of squalene. The basic skeleton arises from the cyclization of 3S-2,3-epoxy,2,3-squalene. Oleanane is an

example of a pentacyclic triterpènes and testosterone of a steroid. Tetracyclic terpenes and steroids have similar structures but have different biosynthetic pathway.

Steroids contain a ring system of three 6-membered and one 5-membered ring because of the profound biological activities encountered, many natural steroids together with a considerable number of synthetic and semi-synthetic steroidal compounds are employed in medicine (e.g. steroidal saponins, cardioactive glycosides, corticosteroid hormones and mammalian sex hormones).

The pharmaceutical applications of triterpenes and steroids are considerable. Cardiac glycosides have been used in medicine without replacement by synthetic drugs. Saponins from ginseng and liquorice exhibit many therapeutic effects. (iv) *Saponins* 

Saponins constitute a vast group of glycosides, which occur in many plants. They are characterized by their surfactant properties; they dissolve in water and when shaken, form a foamy solution. Saponins are classified by their aglycone structure into triterpenoids and steroid saponins; most triterpenoid saponins are derivatives of one of the triterpènes oleanane, ursane and lupane, while steroid saponins generally possess the typical steroid skeleton with 2 extra rings E, a furan structure and F, a pyran structure respectively.

Many saponins have haemolytic properties and are toxic to cold-blooded animals especially fish. The steroidal saponins are important precursors for steroid drugs including anti-inflammatory agents, androgens, oestrogens and progestins. Well-known steroid sapogenins are diosgenin from *Dioscorea*, hecogenin from *Agave* and smilagenin from *Smilax*. Triterpene saponins exhibit various pharmacological activities: anti-inflammatory, molluscicidal, anti-tussive, expectorant, analgesic and cytotoxic. Examples include ginsenosides, which are responsible for some or the pharmacological activity or ginseng and the active triterpenoid saponins from liquorice.



## Cardiac glycosides

The aglycone part of cardiac glycosides is a tetracyclic steroid with an attached unsaturated lactone ring that may have 5 or 6 members. Cardiac glycosides are classified into two groups according to the lactone ring: the C<sub>23</sub> cardenolides with an  $\alpha$ , $\beta$ -unsaturated d- $\gamma$ -lactone (= butenolide), and the C<sub>24</sub> bufadienolides with a di-unsaturated  $\gamma$ -lactone (= pentadienolide). The sugar moiety is normally attached via the C-3 hydroxyl group or the aglycone. The majority or the saccharides found in

cardiac glycosides are highly specific. They are 2,6-dideoxyhexoses, such as D-digitoxose, L-oleandrose or D-diginose. These sugars give a positive reaction with the Keller-Killiani reagent.

Cardiac glycosides have been used as drugs for the treatment of cardiac insufficiency. An example is digitoxin from *Digitalis*, where the sugar moiety is attached to the agycone digitoxigenin via the C-3 hydroxyl group (Padua et al., 1999).



Digitoxigenin

# Carotenoids

Carotenoids contain 8 isoprene (C<sub>40</sub>) units that are responsible for the orange and yellow colours of some vegetables and fruits. Among these compounds, the hydrocarbons are collectively referred to as carotenes and the hydroxylated derivatives as xanthophylls. Carotenoids are either acyclic (e.g. lycopene) or comprise of one or two pentacyclic or hexacyclic rings at one end or the other (e.g.  $\beta,\psi$ -carotene) or at both ends (e.g.  $\beta,\beta$ -carotene).

Carotenoids became interesting agents after the discovery of a negative correlation between the plasma concentration or  $\beta$ -carotene and the prevalence of certain forms or cancer. Some doctors prescribe  $\beta$ -carotene for cancer patients. Furthermore, in the intestine  $\beta$ -carotenes are converted to retinol (Vitamin A). They can be used for the treatment or photosensitization, retinal diseases and glaucoma. Carotenoids are also safe colouring agents for food substances and cosmetics (Padua et al., 1999).

# 14. Biological and pharmacological activity and therapeutic applications

Tastes (sweet, bitter, sour and astringent) are among the many classical examples of biological action of plant materials in man. Other sensations include irritations, itchiness, pungency, acridity as well as the types of euphoria and hallucinations. It is only recently that biological activity is understood in terms of molecular interactions. Plant and plant constituents have a key position in the advancement of modern studies and knowledge on biological activity or substances. There are several reasons for this:

- Firstly, plant species whether traditionally used or not, continue to be important sources or food, medicine and supplementary health products.
- Secondly, the bioactive plant compounds are themselves products (or derived products) of metabolism, and hence function in life processes in a similar way to compounds that operate in humans and animals. Researching hoping to develop drugs from plants need to understand the basics of such functions and mechanisms in relation to the bioactive molecular entities.
- Thirdly, plants also yield products, which are auxiliaries in medicine and pharmacy and sustain or condition pharmacological activity and therapeutic efficacy. In addition, a series or these auxiliary substances are used in biomedical research and in clinical tests.

Testing the biological activity of medicinal or potentially medicinal plant materials demands a special approach. Investigations may be focused on understanding the bioactivity or a compounded plant extract or simply directed at isolating a single bioactive chemical compound. In the latter case, results often lead to oversimplification or wrong explanations or the bioactivity or extract preparations. On the other hand, thorough studies on single bioactive constituents provide important information for plant drug research. However, the much more complex array of molecular interactions and bioactivity mechanisms that arises from plant extracts represents a much greater and more fascinating challenge to science.

# 15. Factors affecting biological activity

Various aspects of bioactivity apply to any chemical whether of natural or synthetic origin. These aspects will be dealt with briefly and are:

## 15.1. Physicochemical properties

These relate to the transport of the bioactive compound to its site of action, usually a receptor or other biomacromolecule at the cellular or subcellular level. Under experimental conditions, either in vivo or clinical, or real life conditions, the extent to which a drug passes through semi-permeable membranes before reaching its site or action depends on its solubility.

Under in vitro conditions, many or these barriers are absent. In vitro bioactivity therefore represents only a stage in this basic assessment of pharmacological effects. In plant drug research, the solubility of active constituents may be revealed from extraction procedures. Extraction programmes separate lipophilic constituents from water-soluble compounds. Further extraction of an extract may lead to further refinement of physicochemical properties.

After the bioactive molecular entity has been identified, detailed data on solubility, partition coefficients and the electrolytic behaviour can be determined. Solubility characters are closely related to drug absorption and the degree of absorption is an important determinant or drug action. Many bioactive plant constituents are weak acids and bases, and their degree or ionization when dissolved, is or great importance to their bioactivity. As a rule, the ionic form is more water-soluble.

These factors are important when bioactivity is regarded in the context of drug distribution between intestine and plasma, between kidney tubules and urine, and between plasma and other body compartments. Generally, but simplified, one may say that only the lipid-soluble and un-dissociated forms of a bioactive molecule will pass through membranes. However, at the site of action, bioactive compounds may generate their action by binding to a receptor on the cell membrane.

# 15.2. Chemical parameters

The structural features of a compound can be related to its pharmacological properties, either qualitatively or quantitatively. The principles, concepts and numerical rules governing qualitative and quantitative relationships between structure and activity help explain the pharmacological activity of a new compound, which is why it is important to evaluate the structure of a newly isolated plant compound. The basic aspects of molecular structures involved in bioactivity include:

- *Resonance and Inductive effect*: This is a phenomenon that a molecule can be represented by two or more structures that differ only in their electron, but not atomic arrangement. The electron density and electron distribution patterns help explain the molecule's activity and hence its molecular interaction and bioactivity. While resonance is explained as at above, inductive effect is measurable electrostatic phenomenon caused by actual electron shiftor displacements along chemical bonds. Both negative and positive inductive effects can lead to a change in bioactivity.
- Oxidative and reductive potentials: This phenomenon represents the tendency of a compound to lose or gain electrons. Electron transfer is vital for living processes and without which living cells would not function. Bioactive compounds derived from plant sources function in enzyme systems in plants in the same way to those in animals or humans.
- *Types of bonding*: The phenomenon of biological activity is concerned with covalent and non-covalent interactions. Covalent bonds are formed enzymatically and are common to all biomolecules. Hydrogen bonds, ionic forces, hydrophobic (or lipophilic bonding), charge-transfer interactions, all representing non-covalent interactions are also common to functional life processes. Some agents can affect physiological functions by forming irreversible covalent bonds with target biomacromolecules and these molecules would be considered to be toxic at cellular level and would be difficult to control clinically and medically. In plant drug research, agents exerting their activity through much weaker and reversible bonding process would be desirable.
- Spatial arrangement of the molecule: In terms of activity, it is important to have a good steric and electronic complementarity between ligand and target biomolecule. Bioactive compounds interact with enzymes by fitting sterically into a binding pocket—the space sterically provided by these targets. Thus the molecular

dimensions, interatomic distances, arrangements of electrons and the stereochemical properties of both ligand and target are decisive in determining biological activity.

# 16. Current status of drug discovery

A recent analysis of natural products as a source of new drugs over the period 1981–2002 shows that 67% or the 877 small molecules, new chemical entities are formally synthetic but 16.4% correspond to synthetic molecules containing a pharmacophore derived directly from natural products. Furthermore, 12% are actually modeled on a natural product inhibitor of the molecule target of interest, or mimic the endogenous substrate or the active site, such as ATP. Thus only 39% or the 877 molecules can be classified as truly synthetic in origin. In the area of the anti-infectives (anti-bacterial, anti-fungal, parasitic, and viral), close to 70% are naturally derived or inspired, while in the cancer treatment area 67% are in this category.

In recent years, there has been a steady decline in the output of the R&D programs of the pharmaceutical industry and the number of new active substances, also known as the new chemical entities hit a 20-year low or 37 in 2001. Further evidence of this drop in productivity is evident from the report of the FDA in 2001, down from 24 the previous year. This downturn has been attributed in part to disruption of laboratory activities by the surge in company merges and acquisitions, the mounting costs of drug development, and the FDA over-caution in the drug approval process.

Recently, there has been rekindling of interest in 'rediscovering natural products'. As stated by one authority "We would not have the top-selling drug class today, the statins; the whole field of angiotensin antagonists and angiotensin-converting enzyme inhibitors; the whole area or immunosuppressives, nor most or the anti-cancer and anti-bacterial drugs. Imagine all of these drugs not being available to physicians or patients today". It is clear that Nature has played and will continue to play, a vital role in the drug discovery process (Cragg and Newmann, 2005).

## 17. Plants used for the endocrine system-diabetes

Diabetes mellitus (DM) is the commonest endocrine disorder that affects more than 100 million people worldwide and in the next 10 years, it may affect about five times more people than it does now (ADA, 1997). History has recorded that as 700–200 BC, this disease was recognized and even distinguished into two types: a genetically based disorder and the other one resulting from dietary indiscretion (Oubre et al., 1997).

This chronic and incurable disease is essentially, caused by lack of insulin, Type 1, or insulin resistance Type 2. Both types are associated with short- and long-term complications that affect the individual's quality of life and often engender fear and powerlessness and can compromise physical and psychological functioning.

Complications are the major cause of morbidity and mortality in DM. Complications arise because of a lack of good blood glucose control. However, achieving good control is difficult for many individuals because the delicate balance hormonal balance that controls glucose homeostasis is disrupted by the disease. Because of the potential threat to quality of life and the chronic nature or diabetes, many people are turning to complementary therapies to assist them to cope and control the disease. Among the complementary therapies, are medicinal plants arising from indigenous medicines.

Indigenous medicine and plants in several cultures, e.g. in India, have always been used for the treatment of DM. Drugs have been derived either directly or indirectly from plants. Some plant products act by lowering the level of glucose in the blood while others act by inhibiting glucose absorption from the gut and hence prevent the surge in blood glucose that can occur immediately after a meal.

The ethno-botanical information reports about 800 plants that may possess antidiabetic potential (Alarcon-Aguilera et al., 1998). Several such herbs have shown anti-diabetic activity when assessed using presently available experimental techniques. A wide array or plant derived active principles representing numerous chemical compounds have demonstrated activity consistent with their possible use in the treatment of NIDDM (Marles and Farnsworth, 1995). Among them are alkaloids, glycosides, galactomannan gum, polysaccharides, peptidoglycans, hypoglycans, guanidine, steroids, carbohydrates, glycopeptides, terpenoids, amino acids and inorganic ions. Even the discovery or widely used hypoglycaemic drug-Metformincame from the traditional approach of using *Galega officinalis*. It can be concluded therefore that plants are a potential source of anti-diabetic drugs but this fact has not gained enough momentum in the scientific community. Several reasons may be advanced for this: lack of belief among the practiotioners or conventional medicine over alternative medicine, alternative forms or medicine are not very well defined, possibly or quacks practicing such medicines providing alluring and magical cures and natural drugs may vary tremendously in content, quality and safety.

Although oral hypoglycaemic agents/insulin are the mainstay of treatment of diabetes and are effective in controlling hyperglycaemia, they have prominent side effects and fail to significantly alter the course of diabetic complications (Rang and Dale, 1991). As the knowledge of the heterogeneity of this disease increases, there is a need to look for more efficacious agents with lesser side effects. Though development or modern medicine resulted in the advent or modern pharmacotherapeutics including insulin, biguanides, pharmacotherapeutics including insulin, biguanides, sulfonylureas and thiazolidinediones, there is still a need to look for new drugs as no drug (except strict glycaemic control with insulin) has been shown to modify the course or diabetic complications.

# 17.1. Hypoglycaemic and anti-diabetic herbs

Several plants have been tested for their anti-diabetic potential. For most of them, the findings have been based on the ethno-botanical claims. The present non-exhaus-

tive list gives an overview of some plants with well-known profiles of anti-diabetic claims.

Aegle marmelos (Rutaceae) (Bael fruit)



This plant originating from India is used against diabetes. In Mauritius, the bark decoction, is drunk by people suffering from diabetes. The tests effected on the aqueous extracts of the root bark, as used by people in India, (1 ml/100gm) showed hypoglycaemic effect which peaked (44%) at 3 h in normal fasted rats. In addition, the same extract completely prevented peak rise or blood sugar at 1 h in OGTT. The hypoglycaemic activity was reduced upon storage of the extract. Aqueous extracts of the leaves (1 mg/kg for 30 days) significantly controlled blood glucose, urea, body weight, liver glycogen and serum cholesterol or alloxanized (60 mg/kg IV) rats as compared to controls and this effect was similar to insulin treatment (Ponnachan et al., 1993). When fed as aqueous leaf extract (1gm/kg/day) to STZ (45 mg/kg IV) diabetic rats for 2 weeks, it decreased malate dehydrogenase levels (an enzyme known to increase in diabetes) in comparison to diabetic controls. The extracts were equi-effective in comparison to insulin in restoring blood glucose and body weight to normal levels (Seema et al., 1996).

It must be reported that aqueous leaf extracts admistered orally for 28 days also normalized STZ (45 mg/kg body weight) induced histo-pathological alterations in the pancreatic and kidney tissues of rats (Das et al., 1996).

# Allium sativum (Liliaceae) (Garlic)

This perennial herb is cultivated almost throughout the world and is used as a food ingredient. Experiments have shown that an oral administration of 0.25gm/kg of ethanol, petroleum ether, ethyl ether extract of *Allium sativum* cause 18.9, 17.9, 26.2% reduction of blood sugar in alloxan-diabetic rabbits (150 mg/kg). Oral administration of 0.25gm/kg allicin (isolated from Garlic) produced hypoglycaemia comparable to tolbutamide in mildly diabetic rabbits (glucose level ranging from 180–300 mg%) while it showed no effect on severely diabetic animals (>350 mg%) (Grover et al., 2002).

Aqueous homogenates of garlic (10 ml/kg/day) administered orally to sucrose fed rabbits (10gm/kg/day) in water for 2 months) significantly increased hepatic glycogen and free amino acid contents, decreased fasting blood sugar, triglyceride levels

in serum, liver and aorta and protein levels in serum and liver in comparison to sucrose controls. It has been shown also that oral feeding or garlic extracts (100 mg/kg) increased cardiovascular functions in STZ rats, prevented abnormality in lipid profile and increased fibrinolytic activities with decreased platelet aggregation. Plasma insulin level increased with concomitant decrease in plasma glucose levels. In addition, daily oral feeding of the same dose for 16 weeks showed anti-atherosclerotic effects in STZ diabetic rats. Thus garlic may prevent diabetic cardiovascular complications (Patumraj et al., 2000).

Aloe barbadensis (Asphodelaceae) (Aloe vera)



This plant is cultivated widely as an ornamental locally but in many countries, Aloe vera is cultivated on commercial scale for its gel and plant extracts. The latter are recommended in Ayurveda for managing painful conditions and it is also mentioned in other Pharmacopeias, namely the Arabic Pharmacopeia, as being useful in managing diabetes.

Extracts of aloe gum effectively increased glucose tolerance in both normal and diabetic rats. Chronic but not single administration of the leaf exudates at a certain dose (500 mg/kg PO) showed significant hypoglycaemic effect in alloxan-diabetic mice.

Nonetheless, single as well as chronic administration of the bitter principle (5 mg/ kg IP) showed significant hypoglycaemic effect in the same model.

The hypoglycaemic effect or single dose of the bitter principle was extended over a period of 24 h with maximum hypoglycaemia observed at 8h while chronic administration (exudates twice daily and the bitter principle once a day for 4 days) showed maximum reduction in plasma glucose level at the 5th day.

Hypoglycaemic effect of aloe and its bitter principle is mediated through the stimulation of synthesis and or/release of insulin from the  $\beta$ -cells of Langerhans (Ajabnoor, 1990).

It has been shown that the dried sap of the plant (half a teaspoonful daily) for 4–14 weeks) has shown significant hypoglycaemic effect both clinically as well as experimentally.

# Catharanthus roseus (Apocynaceae) (Rosy Periwinkle)



Originating from Madagascar but now or wide distribution throughout the tropics, this plant is commonly used in traditional medicine against diabetes. The oral administration of water-soluble fractions and ethanolic extracts of the leaves, have been tested and have been found to show significant dose-dependent reduction in the blood sugar at 4 h by 26.22, 31.39, 35.57 and 33.37% respectively in normal rats. In addition, oral administration or 500 mg/kg 3.5 h before OGTT (10 mg/kg) and 72 h after STZ administration (50 mg/kg IP) in rats showed significant anti-hyperglycaemic effects. No gross behavioural changes and toxic effects were observed up to 4 mg/kg IP (Grover et al., 2002).

## Momordica charantia (Cucurbitaceae) (Karela, Bitter gourd)



The Karela fruit is eaten as a vegetable. The leaf may be made into a tea called 'cerassie'. The juice, extracted from the various plant parts (fruit pulp, seeds, leaves and whole plant), is very common folkore remedy for diabetes. When tested on laboratory animals, *M. charantia* has shown hypoglycaemic as well as anti-hyperglycaemic activity. Polypeptide-p isolated from fruit, seeds and tissue of *M. charantia* showed potent hypoglycaemic effects when administered subcutaneously to gerbils, langurs and humans. The aqueous extracts of *M. charantia* improved OGTT after 8 h in normal mice and reduced hyperglycaemia by 50% after 5 h in STZ diabetic mice. In addition, chronic oral administration of extract to normal mice for 13 days improved OGTT while no significant effect was seen on plasma insulin levels.

Another study carried out recently on *M. charantia* fruit extracts has shown that the latter had a direct impact on transport of fluid in vitro. Everted intestinal sacs from rats mounted in an organ bath containing Kreb solution was used. It was observed that M. charantia extract had a direct impact on water transport with increasing inorganic phosphate concentration with or without D-glucose in the buffer. In the

control experiment, fluid intake was greatly enhanced at high inorganic phosphate concentration (8–10 mM) in the presence of 5.5 mM D-glucose. The addition of 3.0 mg/ml *M. charantia* extract to the serosal side inhibits the uptake of fluid significantly. It has been hypothesized that an increase in inorganic phosphate enhances oxidative phosphorylation thereby increasing the fluid uptake across everted intestinal sacs of rats. This would point to the fact that *M. charantia* extracts reduced fluid absorption capacity and this may be because of interference with the carrier-mediated coupled entrance of glucose and Na<sup>+</sup> across the brush-border membrane (Mahomoodally et al., 2004).

Murraya koenigii (Rutaceae) (Curry leaf, Carripoule)



The Curry leaf is an inevitable ingredient in Indian recipes. It is extensively used as a flavouring agent both in curries and chutney.

It has been shown than an oral feeding of *Murraya koenigii* leaves diet (10% w/w) for 60 days to normal rats showed hypoglycaemic effect associated with increased hepatic glycogen content due to increased glycogenesis and decreased glycogenolysis and gluconeogenesis (Khan et al., 1995). Dietary supplement with curry leaves has been shown to increase lecithin cholesterol acyl transferase activity (Khan et al., 1996).

Curry leaf powder supplementation (12 g providing 2.5 g fibre) for a period of 1 month in 30 NIDDM patients showed reduction in fasting and post-prandial blood sugar levels at 15-day period with no significant changes in serum glycosylated cholesterol fraction, serum lipids, lipoprotein cholesterol levels, uronic acid and total amino acids (Iyer and Mani, 1990).

Ocimum sanctum (Lamiaceae) (Tulsi, Holy Basil)



This herb, considered to be sacred by Hindus, is commonly planted next to temples generally. It is also an ornamental plant and is grown in gardens. The traditional pharmacopeia reports on the use of this plant against diabetes. In 1968, Dhar et al. in Alarcon-Aguilera et al., 1998 reported hypoglycaemic effect of the ethanolic extracts of the leaf. The ethanol (70%) leaves extract or *Ocimum sanctum* has been shown to cause significant reduction of blood glucose level in normal, glucose fed hyperglycaemic and STZ (50 mg/kg IP) induced diabetic rats. This effect was 91.55 and 70.43% of that of Tolbutamide in normal and diabetic rats respectively. Diet containing leaf powder (1%) fed to normal and diabetic rats for 1 month significantly reduced fasting blood sugar, uronic acid, total amino acids, total cholesterol, triglycerides and total lipids (Rai et al., 1997). This plant has also demonstrated anti-oxidant and hypolipidemic effect (Kelm et al., 2000).

Syzygium cuminii (Syn. Eugenia jambolana) (Myrtaceae) (Jamblon, Java plum)



Syzygium cuminii (Leaves and friuts)

This herb, widely distributed throughout India and Africa, is commonly used against diabetes. The decoction of the dried leaves and bark as well as the seeds, have shown hypoglycaemic effect. Oral feeding of *S. cuminii* (170, 240, 510 mg/rat for 15 days) caused 50% reduction of blood glucose or normal fasted rats while chlopropamide showed 52% reduction. In addition, there was a 2.4, 6.8-fold and 9.2-fold increase in cathepsin B activity (proteolytic conversion or pro-insulin to insulin) by plant extract and chlorpropaide respectively (Bansal et al., 1981). Oral administration of the fruit pulp extract to normoglycemic and STZ induced diabetic rats showed hypoglycaemic activity in 30 min possibly mediated by insulin secretion. In addition, the extract inhibited insulinase activity from the liver and kidney. Oral administration or the aqueous extract or the seeds (2.5 and 5.0 mg/kg for 6 weeks) showed hypoglycaemic (>glibenclamide) and anti-oxidant activity.

Daily administration of lyophilized powder of *E. jambolana* (200 mg/kg) showed maximum reduction or 73.51, 55.62 and 48.81 as compared to their basal values in mild (plasma sugar > 180 mg/dl, duration 21 days), moderate (plasma sugar > 280 mg/dl, duration 120 days) and severe (plasma sugar > 400 mg/dl, duration 60 days) diabetic rats. In addition, the treatment also partially restored altered hepatic and skeletal muscle glycogen content and hepatic glucokinase, hexokinase, glucose-6-phosphate and phosphofructokinase levels (Grover et al., 2000).

Trigonella foenum-graecum (Apiaceae) (Fenugreek seeds)



This plant is a very commonly used herb in Indian cooking. It is also popular in traditional medicine as a hypoglycaemic agent. This hypoglycaemic effect of fenugreek seeds has been demonstrated in experimentally induced diabetic rats, dogs, mice and healthy volunteers (both IDDM and NIDDM) (Alarcon-Aguilera et al., 1998).

The isolated fibres, saponins and other proteins from the seeds were given with meals for 21 days to alloxan-diabetic dogs. Significant anti-hyperglycaemic, antiglycosuric effect along with reduction in high plasma glucagons and somastatin (Ribes et al., 1986) were observed.

Oral administration of 2 and 8 g/kg of plant extract produced fall (p < 0.05) in blood glucose both in the normal as well as diabetic rats. 4-Hydroxyisoleucine, a novel amino acid, extracted and purified from fenugreek seeds, has been found to increase glucose-induced insulin release (ranging from 100 µmol to 1 µmol) through a direct effect on the isolated islets of Langerhans in both rats and humans. The pattern of insulin secretion was biphasic, glucose-dependent, occurred in the absence of any change in pancreatic alpha and delta cell activity and without interaction with other agonists or insulin secretion (such as leucine, arginine, tolbutamide, glyceraldehyde).

In clinical trials, administration of fenugreek seed powder (50gm each with lunch and dinner) in insulin-dependent (Type 1) diabetic patient for 10 days significantly reduces fasting blood sugar and improved OGTT along with 54% reduction in glycosuria. In addition, it also showed significant hypolipidemic effect (Sharma et al., 1990).

Erythroxylum macrocarpum (Erythroxylaceae) (Bois de ronde)



The validation of locally used indigenous plants, against diabetes has given some interesting results on the leaf extracts of *E. macrocarpum* of the Erythroxylaceae family. The crude aqueous extract is commonly used in traditional medicine as a diuretic. Experimental findings have shown that 6.0 and 12.0 mg/ml or the crude extract of *E. macrocarpum* had significant inhibitory effect on the absorption, transport and tissue swelling of the rat small intestine. While D-glucose absorption and transport was not significantly affected (p > 0.05), L-tyrosine transport was inhibited at 6–12 mg/ml of the extract. As water molecules (260) are directly coupled to each sugar molecule transported, it was observed that a high concentration of the extract (12 mg/ml), tended to inhibit the transport of glucose across rat everted intestinal sacs, in vitro. It was hypothesized that active phytochemicals such as tannins, phenols and alkaloids in the extract may decrease the permeability of the enterocyte membrane and the energy independent transport of fluid (Mahomoodally PhD thesis—unpublished).

# 18. Plants used in cardiovascular ailments

Among the ills to which flesh is heir is cardiac insufficiency, a condition in which a weakened heart fails to pump enough blood through the body. Cardiovascular disorders are responsible for many deaths throughout the world and this has been attributed to a large extent to a consequence of lifestyle, diet and heredity. Progress has been made when there has been changes in diet, exercise along with treatment with conventional drugs or phytotherapy. Cardiology has benefited greatly with the introduction of many drugs, some of them semi-synthetic based on natural products. Among these compounds one notes the presence of anti-platelet agent (Aspirin), derived from the *Salix* sp., warfarin, an anticoagulant derived from dicoumarol. Other cardiovascular conditions are arrhythmias (*Rauwolfia*), dropsy or oedema, heart failures (*Digitalis, Crataegus, Strophanthus*), anti-platelets and anti-sclerotic drugs (*Allium* sp.).

# 18.1. Arrhythmias and heart failures

Heartbeat is irregular and fluid collects in the arms, legs and abdomen because the kidney cannot perform their normal function. The swelling is known as dropsy or more formally as oedema. This disease syndrome is not new. Ancient physicians knew of it but because they lacked knowledge of the circulation or blood (discovered by William Harvey in 1628) and information on the function or the kidneys, treatment was limited to usually unsuccessful attempts to reduce oedema with medicines, which increased urine production (diuretic agents).

# Rauwolfia serpentina (Apocynaceae) (Radix Rauwolfiae)

Snake-root (*Rawolfia serpentina*, Apocynaceae) is a small shrub native to India, Sri Lanka and the East Indies that was used locally for mental illness and snakebite, long before it was 'discovered' by Western medicine. More than a thousand years ago, the Indian Rig Veda mentioned snake-root in its verses that describe medicinal plants. In Hindi, it is known as *chandra* or 'moon', a reference to its use for 'moon disease' or 'lunacy'. It is also sold as '*pagal ke dawa*'—the traditional herb for insanity. Indian peasants and medicine men and women knew snake root, named because the twisted, woody roots suggest the form of a snake; this seems to be a coincidental example of the Doctrine of Signatures in which the plant is a true cure for the ailment. The Dutch physician and botanist G.E. Rumpf (1627–1702) had observed that mongoose or weasel, before attacking a snake, fortifies itself by eating the leaves of the *Rauwolfia* plant. This way, it can even resist the deadly bites of the Cobra—hence an antidote to the poisonous bites of poisonous snakes.

Snake-root had been cultivated for medicinal use in tropical India, and the roots are dried and ground into a powder that contains more than 60 alkaloids out of which Reserpine and Rescinnamine are among the principal hypotensive alkaloids.



Reserpine

In 1954, concerned that the Snakeroot might be exterminated by over-collection, Indian government officials imposed a brief embargo on its export. Several drugs companies initiated action to cultivate the plant. Exploration in Africa revealed that the related species *R. vomitoria* had high concentrations of Reserpine and it is now the major commercial source of the alkaloid. A third species, *R. canescens* is also used in West Africa where it is also used to treat high blood pressure.

Experimental pharmacology on small animals has shown that powdered Radix Rauwolfiae has shown to lower the blood pressure by various routes of administration. The major alkaloids lower high blood pressure by depleting tissue stores or catecholamines (epinephrine and norepinephrine) from peripheral sites. By contrast, their sedative and tranquilising properties are thought to be related to the depletion of catecholamines and serotonin (5-hydroxytryptamine) from the brain. Following absorption from the gastro-intestinal tract, the active alkaloids concentrate in tissues with high lipid content. They also pass the blood-brain barrier and the placenta. Radix Rauwolfiae products are characterized by slow onset or action and sustained effect. Both the cardiovascular and central nervous system effects may persist following withdrawal or the drug. The active alkaloids are metabolized by the liver to inactive compounds, that are excreted primarily in the urine.

# Toxicity

Radix Rauwolfiae are contraindicated in patients who have previously demonstrated hypersensitivity to the plant or its alkaloids, also those patients with a history of mental depression, during or shortly after therapy with monoamine oxidase inhibitor. Radix Rauwolfiae is known to interact with or be potentiated by some drugs (WHO, 1999).

The use of Reserpine to treat mental illness has now been eclipsed by synthetic drugs but its discovery by Western physicians remains an important milestone in the history of medicine.

# 18.2. Heart failure, dropsy or oedema

Crataegus monogyna (Rosaceae) (Folium cum Flore Crataegi)

This plant of the Rosaceae family is commonly known as the 'Aubepine' is a thorny shrub common to temperate areas or the northern hemisphere, including areas of North America, parts of South America, East Asia and Europe. The aerial parts of the plant including the flowers are used in traditional medicine in the treatment or asthma, to support cardiac and circulatory functions. The main constituents of the fruit are reported to be flavonoids including hyperoside, vitexin-4-rhamnoside. Other phytochemical present are proanthocyanidins, flavonol glycosides mainly in the form of hyperoside, spiraeoside, and rutin. Epi-catechin (epi-catechol) and related proanthocyanidins, phenolic acids are also present. The other characteristic component is Crataegolic acid. The active constituents have not been identified but it is thought that the therapeutic effects are due to the presence of a mixture of them together.

Clinical and pharmacological data indicate that the standardized extracts of Folium cum Flore Crataegi, standardized to 4–30 mg or flavonoids, increase myocardial performance, myocardial circulatory perfusion and tolerance in case of oxygen deficiency, have anti-arrhythmic effects and reduce afterload. Positive therapeutic effects have also been observed on patients suffering from congestive heart failure (Degenring et al., 2003), hypertension, tachycardia and arrhythmia. The procyanidins inhibit angiotensin-converting enzyme.

Although improvements were seen, no long-term trials have assessed the effects of Folium Cum Flore Crataegi on the mortality rates in patients with chronic congestive heart failure.

# 18.3. Venous insufficiency

Venous circulation is one of the many problems encountered by patients suffering from cardiac-related ailments. Plant drugs with anti-inflammatory, antioxidant activity can bring relief to conditions like haemorrhoids, varicose veins and other conditions that involve a better flow of blood. The anti-inflammatory activities are often attributed to the presence of saponins while the antioxidant activity attributed to the presence of flavonoids and other molecules having antioxidant activities. Among the plants that have contributed shown prominence are: Horse chestnut (*Aesculus castanea*) and Gingko (*Gingko biloba*).

Aesculus castanea (Hippocastanea) (Semen Hippocastani)

A.castanea is commonly known as the Horse chestnut. This tree, indigenous to western Asia, is commonly grown in many gardens and parks in Western Europe and in the United States of America. Traditionally, the dried ripe seeds have been used in the treatment or coronary heart disease. The main constituents are the triterpene saponins (up to 10%) and are collectively known as Aescin ( $\alpha$ -aescin,  $\beta$ -aescin and cryptoaescin) and they are considered to be the major therapeutic principles of the seeds.  $\beta$ -Aescin is a mixture of more than 30 different glycosides derived from the triterpene aglycones protoaescigenin (also known as protoescigenin) and barrintogenol C. Other constituents present include the flavonoids (e.g. quercetin, kaemferol and their glycosyl derivatives).



Hydroalcoholic extracts of the seeds have been tested in canine saphenous veins in vitro, and an intravenous bolus (25–30 mg) increased venous pressure in perfused canine saphenous veins, in vivo. Placebo-controlled clinical trials have been carried out to assess the efficacy of oral administration of standardized extracts (250–600 mg) equivalent to 100–150 mg aescin daily, in the treatment of Chronic Venous Insufficiency (CVI). Clinical studies have shown symptomatic improvement in skin colour, venous prominence, oedemas etc in treated patients. Pregnancy-related varicose veins in women, swollen legs during long (15 h) flights also responded positively to treatment. Double-blind placebo-controlled in healthy volunteers also showed improvement in capillary- resistance (WHO, 2002).

## 18.4. Anti-platelet and anti-sclerotic drugs

# Allium sativum (Liliaceae) Garlic

This perennial bulbous herb has been used since time immemorial as a culinary herb. It is particularly notorious because of its characteristic and persistent pungent smell and acrid taste. This is due to the number of sulphur compounds and the main one being alliin. The latter undergoes enzymatic hydrolysis by alliinase to produce allicin when the garlic pod is crushed. Allicin forms a wide range or compounds such as allyl methyl trisulphide, diallyldisulphide, ajoene and many others, which are volatile. Garlic has also been used in traditional medicine to treat asthma, bronchitis, as an expectorant, aphrodisiac, anthelmintic, anti-fungal and also to 'thin the blood'. Experimental pharmacology has shown that the essential oil, water and ethanol extracts of the garlic bulb extract exhibits a wide range of anti-bacterial and anti-fungal activity against a wide range or pathogens (WHO, 1999). The antimicrobial and anthelminitic activities have been attributed to the presence of allicin. Ajoene and diallyl trisulphide also have anti-bacterial and anti-fungal activities.



Several forms of standardized garlic extracts are available on the market but it is known that the sulphur-containing compounds are those that are responsible for the activity.

The properties for which garlic (both essential oil and extracts) is also well known for, are its ability to lower cholesterol and plasma lipids, lipid metabolism and atherogenesis, both in vitro and in vivo. Anti-hypercholesterolaemic and anti-hyperlipidaemic effects have been observed in various animal models after administration of garlic extracts. Clinical studies on serum lipids and lipoproteins reviewed 25 randomized controlled trials, with daily doses or garlic over a period or 12 weeks showed a 12% average reduction or the total cholesterol and 13% in serum triglycerides. Meta analysis of the clinical studies confirmed the lipid-lowering and cholesterol actions of garlic.

During the administration or garlic extract, an increase in fibrinolytic activity of patients suffering from atherosclerosis. Clinical studies have demonstrated that garlic activates endogenous fibrinolysis. Randomized, double-blind and placebo-controlled, cross-over trials of garlic extracts have shown that they significantly increased the mean diameter or the arterioles and venules as opposed to the controls.

The acute and chronic effects of garlic on fibrinolysis and platelet aggregation were also studied in randomized and placebo-controlled experiments. A daily dose of 600–900 mg or garlic powder for 14 days significantly inhibited platelet aggregation, as compared with placebo groups.

Allicin is also an antioxidant and garlic extract protect endothelial cells from oxidized LDL damage. Diallyl sulphide is also thought to inhibit carcinogen activation via cytochrome P450-mediated oxidative metabolism and an epidemiological study carried out has shown that a diet rich in garlic reduces the incidence of cancer.

## 19. Plants used against problems of the CNS

During the history of Mankind, drugs affecting the Central Nervous System (CNS) have focused essentially on those that bring relief to psychiatric disorders. Recently, a lot of focus has been made on those likely to bring relief to those acting on Parkinsonism and epilepsy and more potent analgesics etc. Drugs of plant origin are important in all these areas although not usually for self-medication. Reserpine has been a classical example where this anti-psychotic drug has revolutionized the treatment of schizophrenia and has enabled patients to avoid hospitalization before the introduction of drugs such as chlorpromazine and olanzines and risperidone. Reserpine in the meantime has shown some side effects in depleting the neurotransmitter levels in the brain thus causing severe depression and has recently been involved in the development of breast cancer.

For milder psychiatric conditions, phyto-therapy can still provide support when one takes into account the statistics whereby depression and anxiety still affects one in six persons and that 40% or the people having mental problems will also develop symptoms of anxiety and depression. The latter is more prevalent in women than in men with associated problems like sleep disturbances etc. It is in this context again that phytotherapy is called upon to re-establish a regular pattern of sleep.

Migraines, dementia, Alzheimer disease are many of the problems associated with the CNS, which are being addressed by plant extracts.

# 19.1. Hypnotics and sedatives

It has been reported that the difference between a sedative and a hypnotic agent depends on the dose. Plant products used in this way are not as potent as synthetic drugs but they do not have as many disadvantages as their synthetic counter parts, which are often recommended for short-term use.

Valeriana officinalis (Valerianaceae) (Radix Valerianae).

This plant has a long history in traditional medicine as a digestive aid, and as adjuvant in spasmolytic states of smooth muscle and gastrointestinal pains of nervous origin. It has also been used to treat epilepsy, gum sores, headaches, nausea etc. This herbaceous plant is being cultivated in many European countries, in the US and also in Japan. The parts used pharmaceutically are the root, rhizome and stolons.

Valerian has a characteristic smell, usually described as unpleasant and is attributed to the presence of iridoid valepotriate constituents and other volatile oils. The main components of the volatile oils are monoterpenes and sesquiterpenes including valeranone, valerianol, valerenol, valerenal and valerenic acid and derivatives. Among the valepotriate compounds are: valtrate, didrovaltrate and isovaltrate, which are highly unstable decomposing readily upon storage. The extracts of *Valeriana officinalis* also contain  $\gamma$ -aminobutyric acid (GABA), glutamine and tyrosine.

The pharmacological properties and clinical efficacy of extracts of V. officinalis are attributed to the valepotriates and valepotriate degradation products. The sedative

effects are due to a mixture or compounds namely valerenal and valerenic acid which are both constituents of the volatile oils and also of valepotriate compounds.



Among the modern medicinal uses for Valerian roots are insomnia, stress and anxiety. The sedative activity of *V. officinalis* has been demonstrated in vitro and in vivo. In vitro studies have demonstrated the binding of valerian extracts to GABA, adenosine, barbiturate and benzodiazepine receptors. In vivo studies suggest that the sedative properties of the drug may be due to high concentrations or glutamine in the extract. Glutamine is able to cross the blood-brain barrier where it is taken up by nerve terminals and subsequently metabolized to GABA. Increased GABA concentrations are associated with decreased CNS activity, which may, at least, partly explain valerian's sedative activity (WHO, 1999). Recently several preparations containing valerian root in combination with other herbs (e.g. Hops) reputed to have hypnotic and/or sedative effects have been tested (Abourashed et al., 2004). Nonetheless the required therapeutic dosage, the type or valerian preparations and the optimum period or use for therapeutic effect still needs to be worked out (Diaper and Hindmarch, 2004).

The spasmolytic activity of the valepotriates is principally due to valtrate or dihydrovaltrate. These agents act on centers of the CNS and through direct relaxation or smooth muscles, apparently by modulating  $Ca^{2+}$  entry into the cells or by binding to smooth muscle.

Although the extracts have been clearly shown to depress CNS activity, the identity of the active constituents still remains controversial. Neither the valepotriates nor the sesquiterpenes valerinic acid and valeranone, nor the volatile oil alone can account for the overall sedative activity of the plant. It has been suggested that the baldrinals, degradation products or the valepotriates may be responsible. It is clear unknown whether the activity of *Valeriana officinalis* resides in one compound, some unknown compounds or the synergistic activity of several compounds.

Piper methysticum (Piperaceae) (Kava kava, Rhizoma Piperis methystici)

In the Pacific Islands, the roots of Kava (*Piper methysticum*) have been chewed for hundreds of years. This small shrub with heart-shaped leaves has a thick woody root, which is fermented to give the famous ceremonial drink, which has been offered to important visitors namely the Queen and even the Pope. This drink is used to induce a relaxed sociable state and nowadays it is used medicinally for its tranquilising properties as well as for other disparate complaints. Kava dietary products have been sold worldwide for the treatment of nervous anxiety, tension and restlessness. Recent reports have showed the potential association of kava usage and liver injuries (Hu et al., 2005).

The main components of kava are the kavalactones (also known as kavapyrones) and include kawain, dihydrokavain, methysticin, yangonin and desmethoxyyangonin and dihydrokawain.



Kawain R = R' = H Methysticin R,R' = O-CH<sub>2</sub>-O

Some of the medicinal uses have been supported by clinical data and these include symptomatic treatment of mild states of anxiety and insomnia due to nervousness, stress and tension. In vitro studies had initially provided conflicting data on receptor interactions of kava extract and isolated kavalactones. Current thinking is that kavalactones potentiate  $GABA_A$  receptor activity. Other receptor binding studies demonstrate no interaction with benzodiazepine receptors. Studies involving laboratory animals given kava extract or purified kavalactones have demonstrated several activities including behavioural effects, analgesic activity, neurological effects, anti-convulsant and antispasmodic and anti-microbial activities. Clinical trials have confirmed the efficacy of kava extracts at relieving anxiety in double-blind and placebo controlled experiments.

Overall the randomized controlled trials involving patients with anxiety have suggested that the kava extracts may be as effective as certain benzodiazepines, although further research is needed to confirm these observations. Recently pharmacological investigations of kava and *Passiflora* extract combination have shown there was a significant decrease of the amphetamine-induced hypermotility and significant prolongation of the sleeping phase induced by subcutaneous injections or barbiturates (Capasso and Sorrentino, 2005).

#### Toxicity

Kava extracts must not be taken for more than the limited period without medical advice. Nonetheless patients have been reported to complain about allergic reactions, hepatotoxicity, discoloration of the skin amongst some or the symptoms.

# Hypericum perforatum (Hypericaceae) (St. John's Wort, Herba Hyperici)

This plant has had a long history of medicinal use. This perennial, herbaceous plant native to Europe and Asia has been used traditionally as a 'nervine tonic' and eventually in the treatment of nervous disorders. In recent years, herbal preparations containing the aerial parts of St. John's Wort, have been among the top selling herbal preparations.

The active constituents are thought to have been, initially, due to the presence of hypericin as the major anti-depressant component of the drug. Experimentally and clinically, it emerged that hyperforin is a major component required for anti-depressant activity. The plant contains other biologically active constituents such as flavo-noids, which may act in a synergistic manner with the above-mentioned constituents in acting as an anti-depressant.



Although the extracts of St. John's Wort have manifested activity as a depressant, the exact mode of action is unclear. Biochemical and pharmacological studies have shown that the extracts inhibit the synaptosomal uptake of the neurostransmitters, serotonin (5-hydroxytryptamine, 5-HT), dopamine and noradrenaline (nor-epinephrine) and GABA.

Other effects of the extract of *H. perforatum* include the ability to reduce the level or cholesterol in the blood of small animals. The flavonoid-rich extract has been shown to lower the serum triglycerides, total cholesterol and lipoprotein cholesterol as well as slow lipid peroxidation and enhance antioxidant enzyme activity (Zou et al., 2005).

### Toxicity

Extracts of St. John's Wort are usually well-tolerated by patients over a prescribed period of time. In vitro studies of the methanolic extracts show that there is intensive binding with oral contraceptives than the aqueous infusions (Muller et al., 2004). Other studies have shown that the extracts of St. John's wort can interact with anticonvulsants, cyclosporins, digoxin, HIV protease inhibitors, oral contraceptives, selective serotonin reuptake inhibitor etc.

# 20. Plants used against the respiratory systems

Respiratory disorders such as colds, asthma and bronchitis have and can be treated by phytotherapy. For such ailments leading to infections, the recourse to antibiotics is inevitable. Nonetheless, throughout the duration or the colds and flu-bouts, decongestants (Eucalyptus, Mint), broncholytics and expectorants (Thyme, Mint), demulcents (Mallow) all help in providing relief. Nowadays immune system modulators (Echinaceae) are becoming increasingly popular and effective. Asthma is also becoming prevalent in many countries. They are being treated more aggressively by steroids and bronchodilators, although the latter can also be of natural origins (Ephedrine and Theophylline).

Although isolated components of Ephedra (pure Ephedrine e.g.) of pure Pseudoephedrine are contraindicated in the event or asthma, it must be highlighted that Ephedra drugs have a long history of use without apparent side-effects and this feature would be attributable to the presence of other components in the whole plant.

This feature would appear to be a general feature in phyto-therapy where the synergistic properties of other molecules present may affect the performance of the medication.

#### 20.1. Broncho-dilators and decongestants

### Ephedra sinica (Ephedraceae) (Herba Ephedra)

This erect or prostrate leafless shrub is commonly referred to in Chinese as the Ma'Huang and belongs to the Ephedraceae family. It has a wide distribution and Ephedra species have been found in Afghanistan, Central America, China and India and many other regions of the world. It has been used in ancient Chinese medicine, which is now used worldwide. This plant has given to medicine Ephedrine, which has been a very useful decongestant and bronchodilator and also used against asthma.

The main component of Herba Ephedrae is (-)-Ephedrine in concentrations ranging between 40% and 90% of the total alkaloidal fractions. Other components include, amongst others, (+)-pseudoepinephrine, which is now more widely used for respiratory congestion as it has fewer Central Nervous System (CNS) stimulatory properties. Nonetheless, it must be pointed out that not all Ephedra species contain ephedrine or alkaloids.



Herba Ephedrae preparations have been used traditionally in China against asthma, hay fever, as a bronchodilator, sympathomimetic, CNS and cardiac stimulant. Herbalists have used it against urticaria, enuresis, narcolepsy and chronic postural hypotension. Medicinal uses supported by clinical data comprise are nasal decongestion due to hay fever, allergic rhinitis, sinusitis and as a bronchodilator in the treatment of bronchial asthma.

Ephedrine and Pseudoephedrine are potent sympathomimetic drugs that stimulate  $\alpha_1$ ,  $\beta_1$  and  $\beta_2$  adrenoreceptors. Pseudoephedrine's activity is similar to ephedrine but its hypertensive effects and stimulation of the CNS are somewhat lower.

Ephedrine has been prescribed to patients suffering from acute asthma and in chronic cases that require maintenance medication. Just like other sympathomimetics with a-receptor activity, ephedrine causes vasoconstriction and blanching, when applied topically to nasal and pharyngeal mucosal surfaces. Prolonged and continuous uses are likely to cause rebound congestion and chronic rhinitis.

# Toxicology

Ephedrine, on the other hand, excites the sympathetic nervous system, causing vasoconstriction and cardiac stimulation and is longer lasting. It is most likely to increase blood pressure by elevating both systolic and diastolic pressures and pulse rates. Nonetheless, absorption of the ephedrine and pseudoephedrine is slower after ingestion or the herb than of the isolated alkaloids. It must be avoided by hypertensive patients.

Herba Ephedra has received bad press recently as it has been incorporated in products for weight reduction and thermogenesis (fat burning). The safety and effectiveness of these preparations is currently an issue of debate and requires further debate.

# 21. Immuno-stimulants

Medicinal compounds are described as being immuno-stimulants when they stimulate an immune action, usually measured by an increase in the number of immune cells circulating in the blood or by enhanced phagocytosis after there has been inoculation by a pathogen. It is also understood that claims for an extract to be an immuno-stimulant are difficult to substantiate and that large number of claims are needed. Nonetheless some herbs such as Echinacea and Astragalus are being used extensively for the same purpose.

Radix and Herba Echinacea of several species of Echinaceae are widely distributed in North America and have been used traditionally both by Amerindians and settlers since the 19th century. Both the aerial parts and the secondary roots are used against a variety or conditions such as pain, inflammatory skin conditions.

Radix Echinaceae (*E. angustifolia* and *E. pallida*) are known to contain an essential oil (0.2-2%) and echinacoside (0.4-1.7%). Other identified components are cynarin, chicoric acid, alkamides, caffeic acid esters and chlorogenic acid derivatives amongst others. Trace amounts of pyrrolizidine alkaloids (tussilagine and isotussilagine) are also present.



Echinacoside

The claims for the effectiveness or Radix Echinaceae as a stimulator of the immune system are based on over 350 scientific studies, spanning a period of 50 years. Experimental pharmacology data based on in vitro and in vivo studies have documented the activation of an immune response after treatment with Radix Echinaceae extracts. The immunostimulant effect is brought about three mechanisms: activation of phagocytosis, stimulation of the fibroblasts, increasing respiratory activities and causing increased mobility of the leucocytes. Chemically standardized extracts, derived from roots and aerial parts from the various species have been assessed for their phagocytic potential. All ethanolic root extracts increased phagocytosis in vitro.

One placebo-controlled clinical study on patients with infections of the upper respiratory tract have shown significant improvement when they were treated with aqueous-ethnoalic tincture (1:5) at 90 drops/day (900 mg roots). The duration of the illness decreased from 13 to 9.8 days for bacterial infections and from 12.9 to 9.1 days for viral infections (WHO, 1999).

# 22. Cancer drugs from plants

Cancer remains a major obstacle to overall public health and is responsible for one in every four deaths in the US alone. In 2003, the American Cancer Society had estimated that there would be some over one million new cases on invasive cancer diagnosed with over half a million deaths from basal and squamous cell skin cancers.

Plants have a long history of use in the treatment of cancer (Hartwell, 1982), though many or the claims for the efficacy or such treatments should be viewed with some skepticism because cancer, as a specific disease entity, is likely to be poorly defined in terms of folklore and traditional medicine (Cragg et al., 1994).

A contribution towards the chemotherapy of cancer, natural product secondary metabolites from plants and microbes in particular play a very important role in the amelioration of this group of diseases.

In a recent review dealing with an analysis of antineoplastic drugs available in western countries and Japan, of 140 compounds in total, a majority (54%) are either natural products (14%), natural products derivatives (26%) of compounds made by total synthesis, but modeled on natural product leads (14%). Accordingly, there is a considerable scientific and commercial interest in the continuing discovery of new anti-cancer agents from all natural product sources, inclusive of plant secondary metabolites (Kinghorn et al., 2003).

In the search for novel anti-cancer molecules, again the tropical rainforests have been privileged for two reasons:

1. The tropical forests sustain considerable biodiversity and in some of these, more tree species are found in 0.5 km<sup>2</sup> area than all of North America (Burslem et al., 2001).

2. There is justifying concern about the impending loss of tropical species so some of these species may not be available to future generations of natural product drug discovery scientists (Cox, 2000).

It has been estimated that of the total number of higher plants on earth (310,000–422,000), about 120,000 of these are tropical endemic species. Moreover, some biodiversity 'hot spots' with high floristic diversity are undergoing major habitat loss (Pitman and Jorgersen, 2002).

In the 1970s, perhaps one of the significant break-through in the field of anti-cancer drugs comes from the Madagascan Periwinkle.

Catharanthus roseus (Apocynaceae). Madagascar Periwinkle, Vinca Rosea



This plant had been used and patent medicines such as Vinculin in England and Covinca in South Africa have been marketed for diabetes. Experimental injections of a periwinkle extract have been effective in decreasing the symptoms in a group of diabetic adults.

The Periwinkle has a long history of treating a wide variety of diseases and has also been used for centuries, in Europe, West Indies, Indian Ocean Islands against diabetes. Its use as a source of anti-cancer alkaloids arose from its reputation as a cure for diabetes. This traditional use resulted in preliminary laboratory investigations in 1950s. Laboratory animals developed critically low counts of white blood cells leaving them defenseless against infections caused by bacteria. This result showed that one or more of the Periwinkle alkaloids might slow or halt white blood cell production and this is probably the mechanism that evolved in nature to discourage herbivorous predators from eating the Madagascan Periwinkle.

Since then more than 150 alkaloids have been isolated and characterized, a number of which have been found to be indole alkaloids including dimeric and bis-indole alkaloids. Thus bioassay-guided isolation or the extracts of the plant led to the characterization or the active complex alkaloidal compounds: Vincristine and Vinblastine. They have proven to be effective agents against childhood leukaemia, breast cancer and Hodgkin's disease (a cancer of the lymph nodes), choriocarcinoma respectively.



Vincristine, R = CHO Vinblastine, R = CH<sub>3</sub>

The level of Vincristine in the plant is extremely low (0.0002%) and hence making it a very expensive anti-tumour agent.

Vincristine and Vinblastine exert their anti-cancer properties by inhibiting mitosis by binding to tubulin, thus preventing the cell from making spindles it needs to be able to move its chromosomes around as it divides. Vinblastine is marketed as Velbe by Eli Lilly and is useful in treating Hodgkin's disease, lymphomas, advanced testicular cancer and breast cancers as well as Kaposi sarcoma. Nonetheless, the side effects are significant and include hair loss, nausea, lowered blood cell counts etc. Vincristine is marketed as Oncovin by Eli Lilly and is used to treat acute leukaemia, Hodgkin's disease and other lymphomas. The semi-synthetic vinca alkaloids—*Vindesin* (marketed under the name Eldisine), used to treat leukaemia and lung cancers while *Vinorelbine* (marketed as Navelbine) by GlaxoSmithKline is used to treat ovarian cancer. Vinorelbine has a wider range or anti-tumour activity than the other vinca alkaloids and is used, in combination with cisplastin, in the treatment or patients with non-small-cell lung cancer (Heinrich et al., 2004).

#### *Podophyllum peltatum* (Berberidaceae) (May Apple)

*Podophyllum peltatum* is commonly known as the Devil's apple or Mayapple. This perennial plant is found growing in the woodland in Northern America. The rhizome, which is the most important part of the plant, is known to be toxic. The main components found therein are podophyllotoxin and  $\alpha$ - and  $\beta$ -peltatin, all being toxic.

The lignan, Podophyllotoxin is also found in other species of Podophyllum. These plants have a long history as a medicine, among native North American and Asian tribes (Hartwell, 1982).. They used to gather the rhizomes in the autumn, dry them and grind them to a powder. They would eat or drink a brew of the powder as a laxa-tive or to get rid or intestinal worms. Currently, the extracts are applied on genital warts and some skin cancers. Nonetheless, the extracts and compounds present therein are too toxic to attempt self-medication.

Podophyllotoxin was first isolated in 1880, the structure proposed in 1932. A close inspection of the structures of the active compounds belonging to these classes or compounds has revealed that the presence of some structural features e.g. a 5-membered lactone ring, a 3,4,5,-trimethoxyphenyl group and a methylenedioxyphenyl group, are responsible for their activity (MacRae and Towers, 1984). This natural compound has been used to generate semi-synthetic derivatives namely—Etoposide and Teniposide.

Etoposide is now marketed as Vepesid for small cell lung cancer, testicular cancer and lymphomas while Teniposide is also used in treating brain tumours.



Podophyllotoxin

The mode of action of Podophyllotoxin is that it binds to tubulin and is a member of 'spindle poison' group of agents and functions by preventing microtubule formation. Etoposide and Teniposide work via different mechanisms by inhibiting the enzyme topoisomerase II thus preventing DNA synthesis and hence replication. The difference in mechanism is attributed to the presence of small changes in the stereochemistry of the molecules.

## Taxus brevifolia (Taxaceae) (Pacific Yew tree)

Another example of plant-derived anti-cancer drug is paclitaxel more commonly known by its trademark name—Taxol. Taxol, a complex terpene-based molecule is derived from the Pacific Yew (*Taxus brevifolia*) and is both a generic and a brandname.

The Taxol story started in the 1960s, when a project was undertaken by the National Cancer Institute (NCI), which was involved in the collection of a number of plants to be assessed for their anti-tumour activity. One of them being the very slow-growing Pacific Yew, *Taxus brevifolia*. Extracts of the Pacific Yew were found to stop the growth of several mouse tumours, a case in which ethnobotany provided no clues. Nonetheless although, native Americans did not use the trees specifically for cancers or tumours, an early ethnobotany reference noted that the Bella Coola tribe of British Columbia used Pacific Yew for lung ailments. This reference may have overshadowed the present as paclitaxel is now being used to treat lung cancers that do not respond to other therapies.



Paclitaxel (Taxol)

Paclitaxel, along with several key precursors (the baccatins) occur in the leaves (albeit in very low yields: 0.004% from 12 kg of plant material) of various Taxus species, and the ready semi-synthetic conversions of the relatively abundant baccatins to paclitaxel and active paclitaxel analogs, such as docetaxel (Cortes and Pazdur, 1995) has provided major, renewable natural source of this important class or drugs.

Cancer exhibit un-controlled cell division and paclitaxel stops malignant tumours from growing, by interfering with the micro-tubules that are responsible for dividing the chromosomes during cell division. The microtubules do not disassemble after cell division is complete, and so many microtubules accumulate in the cytoplasm that cell division cease. Paclitaxel inhibits the separation of the tubulin molecules—the protein subunits that compose the microtubules—providing a unique method of interfering with cancerous growth. Clinical trials during the 1980s revealed that paclitaxel could help in 30% of the advanced cases of ovarian cancer and the drug shows promise for other malignancies as well namely melanoma cell lines.

The activity of Taxol, its unusual and novel structure and the fact that it worked by a new mechanism, encouraged further research into this agent. This has resulted in the clinical trials and the development of analogues such as taxoteres. Alternatives for getting significant amounts of this drug was envisaged seriously. The supply issue was overcome with semi-synthesis whereby metabolites present in larger amounts (e.g. 10-deacetylbaccatin III) in the needles of the related English Yew (*Taxus baccata*). This was a significant breakthough as the needles are a renewable resource and hence there was no need to destroy the trees by the removal of the bark especially as a tree can take up to 100 years to give a trunk of 4 inches in diameter.

Research has also produced synthetic paclitaxel along with several similar analogs like Docetaxel, which was marketed as Taxotere by Rhone-Poulenc-Rorer. The latter may prove to be more effective in anti-tumour activity than paclitaxel itself.

The cost of synthetic drugs being high and coupled witht the fact that the Pacific Yew is a slow growing species, this combination proved to be a challenge to the further development of Taxol. Then a chemist discovered a parasitic fungus growing on the Yew bark and found to produce small amounts of paclitaxel even after the removal from its host tree.

Paclitaxel production continued in the fungus even after several generations were grown in the laboratory. It appeared that the fungus acquired copies of the genes for producing paclitaxel from its host tree. Gene transfer from organism to organism does occasionally occur in nature and paclitaxel may benefit the fungus by inhibiting the growth of competing microorganisms.

Fungi grow rapidly and are easily cultured in large batches, this fungus now named *Taxomyces andreanae* (after its discoverer) may prove to be a better source of Taxol than either the Yew tree or synthesis. Altering the growth conditions of the fungus or changing the fungus genetically may eventually result in paclitaxel that are sufficient for all patients and also protecting the Pacific Yew and its habitat from destruction.

Anti-drug discovery is targeting tubulin depolymerising protein as the major target. As a result antimitotic agents constitute an important class of the current anticancer drugs. Hundreds of tubulin inhibitors naturally occurring, semi-synthetic or synthetic, have been reported.

Combretum caffrum (Combretaceae) (African Bush Willow)

The roots of the African Bush Willow (*Combretum caffrum*) from the Southern Africa region is commonly used in traditional medicine against body pain (Neuwinger, 2000).

Screening of the plant extracts have led to the isolation or the Combrestatins. Among these, combrestatins A-4 (CA4), isolated from the South African tree *Combretum caffrum*, is one or the most potent anti-mitotic agents. The Combrestatins are a family of stilbenes, which act as anti-angionic agents causing vascular shutdown in tumours (Holwell et al., 2002) and resulting in tumour necrosis when tested against solid tumours (Cirla and Mann, 2003).

CA-4 shows strong cytotoxicity against a variety of cancer cells, including multidrug resistant cancer cell lines. It has also been demonstrated to exert highly selective effects in proliferating endothelial cells. CA-4 disodium phosphate (CA4DP), a water-soluble pro-drug of CA-4 shows potent anti-vascular and anti-tumour effects in a wide variety or preclinical tumour models (Nam, 2003).

Combrestatin A4 phosphate has undergone successful Phase I clinical trial and is currently in Phase II and has also exhibited the absence of cumulative toxicity (Young and Chaplin, 2004). This has led to a significant number of compounds based upon the combrestatins skeleton to have been synthesized in the search for more effective anti-cancer agents (Li and Sham, 2002). CA4P also displayed significant cytotoxicity against ATC cell lines when compared to paclitaxel and these effects were longer lasting in two cell lines when compared to that of paclitaxel (Dziba et al., 2002).



**Combrestatin A4 phosphate** 

Camptotheca acuminata (Nyssaceae) (Xi Shu or Happy Tree)

The National Cancer Institute, in its search for anti-cancer agents had screened extracts of the woodbark of a native Chinese ornamental tree, *Camptotheca acuminata*, locally known as the 'Happy Tree'.

Initial screening showed that the extracts were found to be active against mouse leukaemia assays. Bioassay-guided isolation led to the isolation of the active agent the quinoline alkaloid—Camptothecin. This compound as its sodium salt, proved to be extremely active against leukaemia cells and solid tumour inhibitions. The molecule has given rise to a host of other anti-cancer drugs including Topotecan, 9aminocamptothecin and CPT-11. Camptothecin and these analogues have been investigated to treat a wide variety of cancers but the compounds are quite toxic. Among the other Camptotheca metabolites, is 10-hydroxycamptothecin, which has been found to be more active than Camptothecin.



R<sub>1</sub> = H, Camptothecin R<sub>1</sub> = OH, 10-Hydroxycamptothecin

Interest in camptothecin grew as a result of its ability to inhibit topoisomerase I, which is the enzyme involved in many important cellular processes by interacting with DNA (Oberlies and Kroll, 2004). With the structure of Camptothecin acting as template, several products have been developed namely topotecan (hycamptamine) and irinotecan (CPT-11) (Friedman et al., 2003).

While camptothecin (as its sodium salt) was in clinical trials at the NCI in the 1970s but it had to be dropped because of severe bladder toxicity. Nonetheless, Irinotecan, being much less toxic than camptothecin, was approved in the US against metastatic colorectal cancer and leukaemias. Irinotecan has much greater water solubility and is a prodrug, being metabolized in vivo by hydrolysis to give topoisomerase I inhibitor which is 1000 times more active than the parent compound. Topotecan has also been approved in the US for ovarian cancer and has been tested also among paediatric patients with resistant and recurrent solid tumours (Martinez et al., 2003).

With the increasing market demand for Camptothecin (estimated at 1 billion in 2003), there has been concerned about the sustainable practice of extraction of the active metabolites from the bark and the seeds. Nowadays, the development of hairy root cultures and the cloning and characterization or genes encoding key enzymes of the pathway leading to Camptothecin formation in plants has opened new possibilities to propose alternative and more sustainable production systems for this important alkaloid (Lorence and Nessler, 2004).

Brucea antidysenterica (Simaroubaceae)

*B. antidysenterica* is a plant growing in the North Eastern part of Africa, particularly in Ethiopia. This plant has been used by the local population against infectious diseases namely dysentery—hence its botanical name. Further search into this plant has resulted in the isolation of the Quassinoid—Bruceantin. The purified compound along with other related quassinoids were found to be toxic to *Entamoeba histolytical*, in vitro (IC<sub>50</sub> = 0.018 µg/ml) (Gillin et al., 1982).



Bruceantin

Further testings were also being carried out for their anti-tumour properties. This led to the isolation of the quassinoid glucosides Bruceantinoside-A and B (Okano et al., 1981, 1985) and bruceanic acids (Toyota et al., 1990). Preliminary cytotoxic tests effected showed the compounds to be active against five human tumour cell lines, malignant melanoma (RPMI-7951), lung carcinoma (A-549), ileocecal adenocarcinoma (HCT-8) epidermo- carcinoma of the nasopharynx (KB) and medulloblastoma (TE-671) and against murine lymphomatic leukaemia (P-388) (Imamura et al., 1993).

In vivo studies using RPMI 8226 human-SCID xenografts demonstrated that bruceantin induced regression in early as well as advanced tumours, and these significant anti-tumour responses were facilitated in the absence of overt toxicity. Apoptosis were significantly elevated in tumours derived from animals treated with bruceantin and it was concluded that bruceantin interfered with the growth or leukaemia, lymphoma and myeloma cells in culture and xenograft models. The clinical efficacy against hematological malignancies is being investigated (Cuendet and Pezzuto, 2004).

## 23. Plants used against infectious diseases

## 23.1. Anti-malarial properties

Throughout Man's troubled history, few diseases have played so tragic a role as malaria. It has killed or incapacitated more people than all plagues, wars and automobiles. More than 10% of the US overseas armies in 1943 had malaria. In 1596, the earl of Cumberland captured Spanish Puerto Rico but could not hold it because his forces were decimated because of malaria. Alexander the Great died of it in June 2323 B.P. Untreated malaria may kill about 1% of those infected. The survivors prone to relapse may suffer from anaemia, weakness, sexual impotence, chronic abortion and secondary infections—all of which lower the value of the individual to self, community, family. Malaria is believed to be the most serious and important parasitic disease in the world. Malaria, nowadays, is the number one infectious disease in the world and over two million people die each year from malaria (Meshnick and Dobson, 2001) and the majority of the victims being young children.

The name malaria was coined in the 17th century by Dr. Francisco Torti by combining the Italian word for 'bad' and 'air' as it has been called the shakes, the fevers, the ague and many other things, none affectionate. Hippocrates had reported several cases of malaria. It was also known even at that time that swamps (Bruce-Chwatt, 1985) and mosquitoes were involved for malaria was rarely found in dry and windy places and disappeared in winter. It was only during the middle of the 18th century that the relationship of mosquitoes to the fevers be accepted. In 1880, the French Physician, Charles Laveran, found microscopic parasites in red blood cells of human victims of malaria. By 1899, the complete life cycle of the parasite, called *Plasmodium* was known. There are four major species of malarial parasites, each causing different clinical types of the disease. The need for mosquito control became obvious and marshes were drained and other control measures were adopted throughout the world.

DDT, in spite of its almost criminal misuse, as an insecticide, saved millions of lives. Even as far back as the 15th century, physicians were dreaming of some medicine, which could cure the disease, what today is referred to as a chemotherapeutic agent—specific for the malarial parasite. One was found in a plant and its history started in Lima, Peru the capital of New Spain.

After Lima was founded in 1520s, it became a proud and wealthy city and where business prospered. The turn-over for business was high at a place where malaria was endemic. The Church fathers noted that the Indians were not so much bothered by the disease and they attributed the cure to the fevers to the bark of a tree which when mixed with water, cured the fevers. The natives called the tree- the Quina or the 'Fever bark tree'. It was the Society of Jesus that recognized the political potential inherent in this powder and soon developed a monopoly on the bark from Peru.

The 'Jesuits bark' was used to cure and convert people. In 1693, the Chinese emperor, the great K'ang His, had bad malarial attack and the Jesuits in attendance at his court, introduced the bark and saved his life; King K'ang His was grateful but he never became a convert.

However by the end of the 17th century, quinine powder no longer Jesuit Powder was the standard treatment for malaria. Spain still controlled trade through its exclusive mandates in Peru and Bolivia. As demand increased, it became obvious that there weren't enough trees available to assure supplies. Bark collectors had to go further and further into the mountainous bush to find the trees, getting lost and dying in the jungles with dysentery and the dart poisons of the Jivaro Indians. In the middle of the 18th century, a group of French botanists had confirmed that there were four species of the tree. This information was later confirmed by Linnaeus who later gave the name Cinchona to the trees to honour the Viceroy of Peru in 1628–1639 (Potier, 2001).

Today, the disease has become very critical and widespread and one of the main reasons for this is that the anti-malarial drugs, including chloroquine, is no longer effective against the disease as its efficacy has been decreased by the spread of the drug-resistant strain. This loss in efficacy has been a major barrier to the effective treatment of malaria and has posed an urgent challenge to discover new anti-malarial drugs. Malaria is caused by four species of the genus Plasmodium, namely P. falciparum, P. vivax, P. ovale, P. malariae. Almost all fatalities are due to P. falciparum infections and therefore the most important species but P. vivax also causes significant morbidity. This shocking reality is largely due to the emergence of drug resistant strains of *Plasmodium falciparum*.

In the early days, quinine was the curative agent for malaria and subsequently, quinoline anti-malarials and related aryl alcohols were developed on the quinine prototype. This led to the emergence or drugs such as chloroquine, and mefloquine. With the rise of parasite-resistant to these anti-malarials, it became a necessity to search for other synthetic and natural product-based agents. Another plant long used in the treatment of fevers in Chinese traditional medicine was considered. The suggestion to investigate wormwood for anti-malarial activity came from Chinese herbal medicine as this herb-qing hao-has been prescribed for fevers by the Chinese physician Li Shi-zen in 1527.

# Artemisia annua (Asteraceae)

Artemisia annua, also known in China as Qinghao, has a long history in Chinese medicine. A. annua or Sweet Wormwood has yielded the agent Artemisinin and derivatives which are potent classes of anti-malarial drugs. The Artemisinins are sesquiterpene lactones and are widely used to treat multidrug-resistant malaria and they act also on cerebral malaria-causing strains of *Plasmodium falciparum*. The clinical efficacy of these drugs is characterized by an almost immediate onset and rapid class reduction of parasitemia (Eckstein-Ludwig et al., 2003; Jung et al., 2004). Artemisinin is now used as an alternative to chloroquinine in areas of China with resistant strains of *Plasmodium* and has been investigated in the United States by the military, since malaria can quickly debilitate troops.

In the meantime, in view of the fact that A. annua gives extremely low yields or Artemisinine (0.01-0.8%), the direct commercialization poses a problem. Therefore the enhanced cell culture of artemisinine either in cell/tissue culture is highly desirable and are being tried (Abdin et al., 2003).

First isolated in 1972, the sesquiterpene endoperoxide Artemisin has been the basis for several semi-synthetic drugs, namely Artemether and Arteether, which have greater solubility in vaccines and greater anti-malarial activity.



Arteether  $R = CH_2CH_3$
Nonetheless while the synthetic and semi-synthetic analogues are being tested, malaria still poses a challenge to poorer countries where these modern anti-malarial drugs are often unavailable. In these countries, randomized pilot trials have been effected so as to investigate the efficacy of traditional tea preparations of *A. annua* in the treatment of uncomplicated malaria. After 7 day medication, cure rates were on average high (74%) but recrudescence was high which suggested that monotherapy with *A. annua* cannot be recommended as alternative to modern anti-malarials (Mueller et al., 2004; Willcox et al., 2004) but a combination with other anti-malarials, is being recommended (Balint, 2001).

The next question that remains is—How quickly malaria will evolve immunity to artemisinin? In fact the best defense would be an effective vaccine.

Quillaja saponaria (Rosaceae) (Soap bark tree)

Several alternatives of vaccine are being worked upon. The extracts from the South American tree, *Quillaja saponaria*, contain triterpenoid saponins (Guo and Kenne, 2000), which are ingredients in an experimental malaria vaccine (Bienzle et al., 2004; Kirk et al., 2004). Partial purification of the crude extract has resulted in the isolation of Quil A, later named Stimulon. Stimulon seems to work as an adjuvant, a pharmacological additive that improves the effectiveness of a vaccine in promoting the formation of antibodies.

While searching for a vaccine, research going on in other parts of the world where malaria is endemic. With the re-emergence of malaria in the central Highlands of Madagascar in the 1980s and coupled with the lack of inappropriate drugs, pushed the population towards traditional herbal remedies among them were the *Strychnos* species.

### Strychnos myrtoides (Loganiaceae)

*Strychnos* species are commonly used in the local pharmacopeia in Madagascar as well as in mainland Africa. The roots have been used against constipation, tooth-ache, coughs as well as epilepsy. The aerial parts also been used been used against malarial fever (Neuwinger, 2000).

With the prevalence of quinine-resistant, *Plasmodium falciparum*, in Madagascar attention has been focused on medicinal plants that could reverse resistance in malaria. Investigation into several plants led to the investigation of *Strychnos myrtoides* as the crude alkaloids were empirically used as an adjuvant to chloroquine in Malagasy herbal remedies. When combined with chloroquine at a dose level lower than their IC<sub>50</sub> value, they markedly enhanced in vitro, the effectiveness of the synthetic drug against a chloroquine-resistant strain of *P. falciparum*. They also enhanced in vivo chloroquine activity against a resistant strain of *P. yoelii*.

This led to the isolation of two major bioactive constituents—Strychnobrasiline and Malagashanine (Rasoanaivo et al., 1994) as well as four minor alkaloids. Malagashanine, turned out to be the parent compound of a new subtype of Strychnos alkaloids, the C-21, Nb-secocuran indole alkaloids, isolated so far from the Malagasy *Strychnos* (Rafatro et al., 2000; Rasoanaivo et al., 2001; Martin et al., 1999).

Strychnobrasiline and Malagashanine were devoid of both intrinsic anti-malarial activity both in vitro ( $IC_{50} = 73.0 \ \mu g/ml$  for strychnobrasilin and  $IC_{50} = 69.1 \ \mu g/ml$ 

for malagashanine and in vivo 10 mg/kg conferred as a 5% suppression of parasetemia) and cytotoxicity (Rasoanaivo et al., 1996) but exhibited significant chloroquine-potentiating actions which could justify the empirical uses of *S. myrtoides*.



At present, the infusion of the stem barks of *S. myrtoides* in association with chloroquine has been successfully evaluated in a clinical setting. The aim now is to develop purified and standardized extracts for clinical trial and to eventually develop efficient and inexpensive drugs for the treatment of chloroquine-resistant malaria (Rasoanaivo et al., 1996).

It has a weak anti-plasmodial action, but when combined to chloroquine at concentrations much lower than required for anti-malarial effect, it enhanced in vitro and in vivo, chloroquine action against chloroquine-resistant strains of *Plasmodium* malaria.

#### Calophyllum sp. and Garcinia sp. (Clusiaceae family)

Recently the xanthones from the extracts of *Calophyllum caledonicum* and *Garcinia vieillardii* (Clusiaceae) have been tested for their anti-malarial activity against the chloroquino-resistant strains of *Plasmodium falciparum*. The most potent xanthones were found to be the following: demethylcalabaxanthone, calothwaitesixanthone and 6-deoxy-gamma-mangostin with an IC value of 1.0microg/ml (Hay et al., 2004).

# 23.2. Plants and AIDS

Worldwide millions of people are infected and are being infected with the Human Immunodeficiency Virus (HIV), the pathogen that causes Acquired Immunodeficiency Syndrome (AIDS). AIDS is a complex array of disorders resulting from the deterioration of the immune system and infected individual become susceptible to rare forms of cancer; common microbes become opportunistic pathogens. HIV uses cells of the immune system (macrophages and helper T cells) as sites for reproduction and multiple copies of the viral genetic material (RNA) are made and package into new viral particles ready for dispersal into new viral hosts. More cells of the immune system are killed or damaged with each round of infection, when millions of viral particles may be produced each day. Despite the production of antibodies and helper T cells that fight the disease, eventually the virus prevails and the infections and cancer associated with AIDS begin to appear. With no known cure or vaccine against HIV, drugs slow the progression of the viral infection and the onset of AIDS. New anti-HIV compounds from natural sources are reported almost daily, some essentially unproven and others with distinct promise based on in vitro research. Secondary metabolites will play a significant role in combating viral infections along with the AIDS infections incurred by a compromised immune system. More than 36,000 extracts have been tested by the National Cancer Institute of the USA and 10% of them have been reported to exhibit some anti-HIV activity.

One of the most promising anti-AIDS compounds is produced by the Malaysian tree, a member of the tropical Garcinia family (Guttiferae-Clusiaceae) that is valued both for its wood and resins. A detailed survey of C. lanigerum and related species showed that latex of Calophyllum teysmanii yielded extracts with significant anti-HIV activity. The active constituent was found to be (-)-calanolide B, which was isolated in yields of 20-30%. Eight compounds have been isolated from C. lagenirum with Calanolide A showing anti-HIV activity and C. teysmanni has yielded Calanolide B, equally found to be slightly less active than the (+)-Calanolide A, but it has the advantage of being readily available from the latex which is tapped in a sustainable manner by making small slash wounds in the bark of mature trees without causing any harm to the trees. Chemically, Calanolide A is a coumarin and is now being tested in human trials. The drugs are being developed by Sarawak Medichem Pharmaceuticals, a joint venture company formed between the Sarawak State Government and Medichem Research, Inc. (+)-Calanolide A (which has been synthesized by Medichem chemists) is currently in Phase II clinical trials, while (-)-calanolide B is in pre-clinical development. These two Calanolides can also be isolated from other *Calophyllum* species, namely from the leaves or *C. brasiliensis* (Huerta-Reyes et al., 2004) and exhibiting more or less the same pattern of activity.



Eventually, it may be one of the antiviral ingredients included in the AIDS 'cocktail' that slowed the rate of AIDS progression and extended the lives of HIV-infected patients.

Another potential anti-HIV drug originated in Africa, comes from the woody vine *Ancistrocladus* species. The crude extract from this plant has yielded Michellamine B a new alkaloid, which in the initial trials have been shown to work against the HIV virus.

Michellamine B is a chemically stable molecule found to be present in the leaves even after the leaves have fallen to the ground.



**Michellamine B** 

Based on the observed activity and the efficient formulation of the di-acetate salt, the NCI committed Michellamine B to advanced pre-clinical development, but continuous infusion studies in dogs indicated that in vivo effective anti-HIV concentrations could only be achieved close to neuro-toxic dose levels. Thus, despite in vitro activity against an impressive range of HIV-1 and HIV-2 strains, the difference between the toxic dose level and the anticipated level required for effective antiviral activity was small, and NCI decided to discontinue further studies aimed at clinical development. However, the discovery of novel anti-malarial agents, the korupensamines, from the same species (Hallock et al., 1994), adds further promise for this species.

#### Homalanthus nutans (Euphorbiaceae) (Mamala)

Still in the search for new anti-AIDS compounds, Prostratin was isolated as the active constituent from an extract of the wood of the tree, *Homalanthus nutans* (Gustafson et al., 1992) growing in Samoa.

This breakthrough came as a curiosity by ethnobotanist Paul Cox who was working in Samoa. He observed that the inner bark of *Homalanthus nutans* was used to treat yellow fever, which is a clinical manifestation of the viral disease hepatitis. He carried out several interviews with traditional healers in Samoa, collected samples and subsequently sent the samples to the NCI for assessment of the antiviral activity in anti-AIDS assay.

Subsequent studies determined that prostratin is a relatively polar 12-deoxyphorbol ester. When it was discovered that the main active compound was a phorbol ester, interest was greatly reduced because it is a known fact that phorbol esters are strong tumour-promotors. Nonetheless, the extracts from this plant were tested for their tumour-promoting ability. It was found that this compound did not promote tumour formation but instead prolonged the life of HIV-infected cells and stops the infection of healthy cells by HIV (Gustafson et al., 1992).

Prostratin is therefore, a potent activator of HIV expression in latently infected T-cell lines (Gulakowski et al., 1997), and its potential value in HIV therapy lies more in its possible utility as a viral activator rather than as an anti-HIV agent.



Several other botanical drugs may be useful in treating AIDS-related infections and cancers. The alkaloid berberine, found in members of the Poppy family (Papaveraceae) has been used to treat infections caused by bacteria, fungi and protozoans. As a broad-spectrum antibiotic with few side effects, berberine has potential for treating the various forms of severe diarrhoea associated with AIDS. Protozoans in particular cause gastrointestinal infections in people with damaged immune systems, and perhaps a maintenance dose of berberine would help to control such opportunistic pathogens.

# Catharanthus roseus (Apocynaceae) (Madagascan Periwinkle)

AIDS patients also find themselves at risk for cancers that are usually controlled by a normal immune system. Vinorelbine, a semi-synthetic version of one anti-cancer alkaloid from the Madagascan Periwinkle (*Catharanthus roseus*, Apocynaceae) disrupts the spindle fibres that are responsible for separating chromosomes during cell division. It works at lower concentrations and with fewer side-effects than the alkaloids derived directly from the plants and it could be useful in combating Kaposi's sarcoma, a rare skin cancer associated with AIDS.

#### 24. Medicinal plants, functional foods and nutraceuticals

## 24.1. The functional food concept

Over the past 15 years, the consumer's interest in healthy nutrition has changed considerably. Earlier, good nutrition meant avoiding products with high calorie, salt and fat content. Today, more attention is being paid to positive/preventive nutrition

owing to the increased choice of food products with the desired functional components and contents. Several reasons can be forwarded as to why this preventive concept has come to prevail. Nutritional research is playing a leading role in emphasing on more preventive and public health aspects. Furthermore, health professionals have learned through positive experience with fortified foods.

Currently, there is no legal definition for functional foods, either in Europe or in the US. Only Japan has introduced an official definition for 'Foods for Specific Health Use' (FOSHU) in 1991. For the Food Industry, the working definition is as follows: 'Functional Foods are foods and beverages with a specific health-promoting effect based on scientific proof'.

In view of a lack of an unequivocal definition, problems arise with consequences for market research and legislation. Confusions about the terms 'nutraceuticals' and 'functional food' arise especially as both terms are being used interchangeably. Functional foods appear to be foods whereas nutraceuticals may be produced from foods but are marketed in concentrated form as pills, capsules, powders, tinctures etc. either as a single of mixed preparations.

In the US, this ambiguity still allows companies to distribute foods containing herbs that do not fulfill the GRAS (Generally Regarded As Safe) status under DSHEA (Dietary Supplement Health Education Act) as herbal supplements. It is agreed nonetheless, that functional food ingredients, need to be GRAS. This statement also covers botanical functional food ingredients. The term 'botanical' is generally used in the broader sense of the word as it would include herbal drugs/extracts and phytochemicals. Phytochemicals are enriched or selected fractions of plant extracts. Currently, the terms are being used for secondary metabolites of plants that may exert health-related effects. Therefore, botanicals are the bioactive ingredients added to or already inherently present in foods and beverages, in order to make it functional.

The key driving forces for the functional food concept has been progress in nutritional science. This coupled with the changes in lifestyle in industrialized countries where people have started to adopt a more active and healthy lifestyles. Added to this are health and educational campaigns as well as commercial advertising. Prevention-oriented medicine and more self-responsibility in health issues are becoming a political necessity, especially with an ageing population. 'Value-addition' to 'regular foods' is gaining popularity rapidly, especially to an industry which has for long remained traditional.

## 24.2. Categories of botanical functional food ingredients

## Antioxidants

Antimutagenic and anticarcinogenic agents Antimicrobial and antiviral substances Enhancers of the gastrointestinal function Immune-modulators and stimulators Inflammation-inhibiting substances Cognitive enhancers (psychrotrophic/neuroregulatory substances) Oestrogen-modulators Blood-pressure-reducing agents Cholesterol-reducing agents Anti-Allergenics Anti-Diabetics

An ingredient may have beneficial effects in more than one category, whereas other react very specifically. Furthermore, scientific evidence for efficacy and more importantly clinical proof is quite heterogenous, both within a category and across categories.

Traditional ingredients (such as soya proteins) have been re-discovered, phytochemicals in fruits and vegetables (e.g. resveratrol) are recent discoveries, as are innovative ingredients (e.g. phytosterols). The complete mode of action has been elucidated only for a few ingredients.

The most widely used, as well as the most promising ingredients categories or plant origin are highlighted and emphasis is also placed on the commercial and marketing aspects. It must be pointed out nonetheless that the functional food industry has focused mainly on those botanical ingredients that are inherent to vegetables, grains and fruit which are part of the normal human diet, their specific nutritional value and or/effective dose that often remained obscure, until recently when epidemiologists/clinical research have shed light on them.

Ethnobotanical studies are also highlighting the applicability of other herbs and plant sources for more potent or even innovative ingredients. So far safety concerns, as well as time-consuming efficacy and toxicological studies have prevented a broader use or herbal ingredients in functional foods.

# 24.3. Traditional 'functional foods'

*Soya proteins*: The functional components of Soya extracts include proteins, isoflavones, oligosaccharides and phospholipids. Soya proteins have been reported to have beneficial effects directed towards the most relevant diseases in industrialized countries, including cardiovascular disease, cancer and diabetes. In Japan, Soya proteins have FOSHU status as an active ingredient in food to reduce blood pressure. In 1999, the FDA released a health claim approval, which stated that: "25 g or soya proteins a day, as a part of a diet low in saturated fat and cholesterol, may reduce the risk of heart disease".

There is a major problem with Soya—especially among European consumers- is its lack or acceptance due to the fact that soya is a typical GM (Genetically Modified) crop. In fact, in 2001, US soya bean farmers planted around 63% of their soya bean field with GM soya and also non-GM alternatives are becoming scarce. Organically grown soya for use as food ingredients is increasingly being imported from South America.

*Phytosterols*: Phytosterols and stanols are currently among the most successful phytochemicals for the development of functional foods with unique health claims. Phytosterols have clinically proven cholesterol-lowering properties.

*Prebiotics*: An alternative approach for dietary modulation or intestinal flora instead or oral administration of living bacteria ("probiotic") isto specifically stimulate the growth of endogenous microorganisms by 'prebiotics'. These are defined as 'nondigestible food ingredients that beneficially affect the host by selectively stimulating the growth and/or activity of one or a limited number of bacteria in the colon, and this improves host health'. Prebiotic are basically non-digestible carbohydrates, mainly obtained by extraction from plants, followed by enzymatic hydrolysis. The fructose-based disaccharide inulin and oligofructose are the most widely used as food ingredients, because of their technical (fat replacement, gelling) and nutritional ('bifidogenic') properties. Both are increasingly used in functional foods, especially in dairy and bakery products. The primary source or inulin is the roots of the Chicory (70% or the dry materials).

*Fibres*: Apart from prebiotic disaccharides inulin and oligofructose, there are other natural oligo- and polysaccharides, including pectins, gums and b-glucan, collectively called soluble dietary fibre, that exert beneficial effects on intestinal function by increasing the physical bulk in the bowel and stimulating intestinal transit. In addition, the functional food concept makes use or the hypocholesterolaemic properties or water-soluble fibres. Soluble fibres are already added to a variety of functional food products such as milk products, bakery goods, confectionary and soft drinks.

Pectin, a structural component of plant cell walls is produced primarily from apple pomace and citrus peel and other by-products of juice manufacture.

Psyllium seed husks had previously been recommended after having received approval for a health claim but commercially it was not successful.

Gums, used in food processing are the exudates Gum Arabic. Guar gum from *Cyamopsis tetragonolobus*, is an annual leguminose grown in India and Pakistan. Increasingly seaweed gums such as Carrageenan (extracted from red seaweed) or algin (brown sea weed) are another source of soluble fibres. Seaweeds are used extensively in Asian kitchen. It contains other interesting components or traditional medicinal value with curative powers for a variety of diseases (tuberculosis, arthritis, colds, influenza, cancer etc.). 'Nori' (*Porphyra* sp.) a red alga popular in Japan and Korea has very high proteins, iodine and vitamin C content. Nori is the most valuable single crop produced by aquaculture worldwide.

*Phytoestrogens*: There are many plants that contain oestrogenic substances (phytoestrogens) and pharmacological and epidemiological evidence suggests that they act as mild oestrogens or, in some instances, can act as anti-oestrogens (in as much as they bind to oestrogen receptors and they prevent the occupation by natural oestrogens).

Phytoestrogens are gaining popularity for the development of functional food products targeted at women suffering from menopausal symptoms and to prevent osteoporosis. They may also reduce the risk or hormone-dependent cancers, such as breast cancer in women and prostate cancer in men. Because of the structural similarity to oestradiol and its binding ability to the human oestrogen receptor, they are suspected to act as natural SERMs (Selective Oestrogenic Receptor Modulators). The three major classes of phytoestrogens are described—isoflavones, lignans and coumestrans. Some species of Palm even contain similar hormones (e.g. estriol). Soya is a particularly rich source of isoflavones, whereas flax has the highest known lignan content.

The common occurrence of these substances, has implications for men as well as for women, in that the incidence of benign prostatic hyperplasia is lower in men, and menopausal symptoms in women, in societies consuming significant amount of foods containing these substances in their normal diets. Although it is known that they have beneficial effects, including chemopreventive activity, the full mechanism of action is not known. It is also worth pointing out that the efficacy and long-term safety need to be investigated more extensively before these purified phytoestrogens are more widely used commercially as functional ingredients.

*Plant oils*: Polyunsaturated fatty acids (PUFAs) are basically divided into 2 families—the Omega-3 and the Omega-6 PUFAs. Both are essential because the human body is not able to synthesize the double bonds in the molecule.

Clues to the risk-reducing properties of omega-3 PUFAs against coronary heart disease point back to epidemiological observations or Eskimos consuming a diet extremely rich in fatty fish. Today, clinical evidence of protective effects of omega-3 PUFAs on conditions such as arteriosclerosis, thrombosis, blood pressure and cardiac function is available. Further omega-3 PUFAs have significant anti-inflammatory properties, making them potentially useful for rheumatoid arthritis and inflammatory bowel diseases. Intake or omega-3 PUFAs has also shown positive effects in the treatment or several psychiatric disorders.

Still, the primary commercial source of omega-3 PUFAs is fish oil. However, vegetable oils and 'single-cells' oils from marine algae (e.g. *Cryphtecodinium cohnii*) and fungi (e.g. *Mortierella alpina*) gain ground because of better acceptance by the consumer. Flax seed and rapeseed oil, dark green leafy vegetable, legumes and nuts all contain considerable amounts of omega-3 PUFAs.

The demand for fish oil alone has been forecast to reach 25,000 tons by 2010, making alternative sourcing mandatory. Fish oil preparations are usually distributed as dietary supplements in the form of capsules.

Omega-6 PUFAs such as  $\gamma$ -linolenic acid (GLA) have pronounced anti-inflammatory properties. The major dietary sources or GLA are seed oils from Borage, Primrose abd Blackcurrant. Although they are currently only marketed as encapsulated supplements, stabilised powders for fortification of foods are already available.

Phospholipids are another interesting group or plant lipid ingredients. The main sources are common seeds such as Soya, Rapeseed and Sunflower. Phospholipids (lecithin) have been primarily used as emulsifiers in the food industry. However, there is now increasing evidence that they can lower elevated serum cholesterol levels and the balance or HDL:LDL ratio. Moreover, phospholipids have been shown to have beneficial effects on age-related memory loss (phosphatidylserine in particular) and functional decline of the immune system (Gruenwald et al., 2002).

# 24.3.1. Vitamins

Vitamins are the only functional food ingredients category that has clear government recommendations. *Herbal supplements*: Herbal supplements is a very important segment for botanical use both in volume and diversity and accounts for about 25% of the total dietary supplements market. It is nonetheless, becoming very difficult to establish boundaries between functional food products and herbal supplements as several enriched food products are marketed as dietary supplements rather than food.

While there has been a decline in the sales values among the top 10 selling supplements (kava kava, gingko, Echinacea, garlic, ginseng, St. John's Wort, Saw Palmetto etc.), there has been some winners (soya, valerian, elderberry, guarana, green tea etc.). This decline may be explained to the negative media coverage about herbs.

# 25. The need for validation

# 25.1. Quality and safety: Production, standardization and quality control

Plant drugs, also known as phytomedicines or phytopharmaceuticals are plant-derived medicines that contain a chemical compound or more usually mixtures of chemical compounds that act individually or in combination on the human body to prevent disorders and to restore or maintain health.

Pure compounds or chemical entities are either isolated from natural products or made by synthesis in the laboratory. Herbal teas, decoction, alcoholic extracts are also traditional ways of using medicinal plants. Very often these plant materials are used in a non-standardised manner. However, nowadays more and more emphasis is being put on the use of standardized materials.

# Doses and efficacy

Two questions have confounded the first person who sampled a medicinal plant— Do herbal remedies work ? and How much should a patient ingest for an effective and safe cure?

If one goes back and reads about the history of Mankind, the battle against diseases in all the cultures of the world, the search for miracle cures for diseases like cancer, malaria and leukaemia etc.., then the answer would be a resounding Yes. However, in many instances, skepticism about the efficacy of quite a few plants is also justified. This skepticism has been growing as a result of the many unrealistic claims made by producer of herbal products.

Scientists have been proceeding in a systematic way in order to validate the claims of several medicinal plants. For some symptoms and ailments, this may be fairly easy to prove but in more complex health conditions, the situation becomes a bit complicated. Nonetheless, medicinal plant extracts are showing a great deal of promise even in instances of complexity of illnesses.

In order to proceed with the validation of the efficacy of medicinal plants, there are several levels of evidence that are taken into account (WHO Monographs, 1999, 2002).

1. The Ethnobotanical claims

2. Anecdotes

- 3. Pharmacological studies
- 4. Observational studies
- 5. Clinical studies

Millions of dollars are spent each year on herbal products that are marketed as food supplements but in reality very few know, chemically, what they are purchasing or using. Very often the dosage varies from the different brands of the same herbal product. Inspite of these major short-comings, there has been a phenomenal increase in the interest towards phytoremedies. The chemistry and efficacy of many of these plants are relatively unknown and there is a chance of toxicity or overdose until the secondary compounds are known and understood.

There is a tendancy in the US and in Europe to regulate and licence this market and this has led to greater and more effective use of these important medicinal plants. There is also general agreement that chemical standardization is the way forward in order for herbal remedies to be prescribed to patients who seek to be treated with medicinal plants.

## 25.2. Standardisation for plant-derived ingredients in medicinal products

*Standardisation* is a method of assuring a minimum level of active ingredients in the extract and is becoming increasingly important as a means of ensuring a consistent supply of high-quality phyto-pharmaceutical products. It can be defined as the establishment of reproducible pharmaceutical quality by comparing a product with established reference substances and by defining minimum amounts of one or several compounds or groups of compounds. In the field of phyto-medicines, standardization only applies to extracts. Standards for active ingredients to be used in medicinal products may be found in monographs and/or pharmacopeias.

#### 25.3. Why is standardization important?

It is accepted that concentration or dosages are very important because herbal medicines (in common with conventional medicines) contain biologically active substances that may produce non-trivial side effects when taken in excessive doses. Very low doses, on the other hand, may have no therapeutic value. In practice, plant material is often highly variable, so that a minimum concentration or a concentration range is often used rather than an exact level. An upper limit is necessary with highly active or potentially harmful ingredients, as most plants have a wide therapeutic window (e.g. a toxic compound is considerably higher than the therapeutic dose). In the case of compounds with a narrow therapeutic window, chemical entities are favoured, as opposed to extracts.

These phyto-drugs when they become registered become a *medicine* that needs to comply with the basic standards required for all drugs.

Standardisation also allows comparison of the clinical effectiveness, pharmacological effects and side effects of a series of products (e.g against a placebo). Standar-

dised products provide more security and increase the level of trust people have in herbal drugs.

At the international level, the World Health Organisation has developed a strategy to review traditional medicines and included within this review is a programme to develop monographs for herbal ingredients (see down below on the Legal Framework). Additionally, the European Scientific Cooperative on Phytotherapy (ESCOP) was established in 1989 to advance the scientific status of phytomedicine and to assist with the harmonization of their regulatory status at the European level. ESCOP has already published 60 monographs on the medicinal uses of plant drugs that have been submitted to regularoty authorities across Europe and accepted by the Working Party on Herbal Medicinal Products of the European Agency for the Evaluation of Medicinal Products (EMEA) as providing the basis for proposed core-SPCs for European decentralized marketing authorizations.

A Pharmacopeia is a collection of quality standards for medicines and their components. In order to obtain marketing authorization for a medical product, the ingredients or the medicinal product must generally comply with a pharmacopeial standard. Thus Pharmacopeial standard may provide guidance on acceptable purity criteria for that ingredient.

# 25.4. Some of the existing Legal Framework for plant-derived ingredients with medicinal properties

## 25.4.1. The World Health Organisation (WHO)

The WHO views herbal medicines as herbs, herbal materials, herbal preparations and finished herbal products that contain active ingredients parts of plants, and other plant materials, or combinations. The WHO recognizes that the traditional use of herbal medicines refers to the long historical use of these medicines and that they may be accepted by national authorities. As a result of this view, the WHO Traditional Medicines Strategy 2001–2005 was developed to review a framework for action for WHO and its partners aimed at enabling TM/CAM to play a far greater role in reducing excess mortality and morbidity especially among impoverished populations.

#### 25.4.2. European Union (EU)

The EU directives 2001/83/EC on the Community code relating to medicinal products for human use lays down a general framework for pharmaceutical products requiring pre-marketing approval before gaining access to the marketand laying down the requirements for the documentation of quality, safety and efficacy, the dossier and expert reports. This framework has effectively been in operation and additionally the European Agency for the Evaluation of Medicinal Plant Products (EMEA), which acts as a central agency for single European medicines marketing authorizations operates a Herbal Medicinal Products Working Party (HMPWP). However, individual Member States (UK, Germany, France, Italy etc.) have taken different approaches in reviewing herbal medicines.

#### 25.4.3. The United States (US)

In the US, the Food and Drug Administration (FDA) has responsibility for food and drug products. Drugs are regarded as products that claim to treat, cure, mitigate or prevent a disease. Herbal medicines follow the same procedures as those for a chemical drug. Otherwise natural products are regulated as foods under a requirement for ingredients tobe generally recognized as safe (GRAS). Natural products generally have GRAS status, provided that this is supported by expert concensus. Hence dietary supplements and herbs are considered to be foods provided that they are generally regarded as safe and do not make medicinal claims.

Furthermore, the ingredients and the plants or parts of the plants must be quantified and where, ingredients are listed with a pharmacopeial reference, they must meet the standard laid down in the pharmacopeia. There are also specific requirements for food additives that do need a pre-market approval by the authority.

## 25.5. Standardised extracts

These extracts as mentioned above are those for which the active constituents (single or groups) are known. They can thus be standardized to be a defined content of the active constituents giving a clearly defined amount of an active natural product. Examples include:

Onion bulbs (Bulbus Allii cepae, Onion)

*Gingko extracts* (Folium Gingko): Standardized extracts (dry extracts from dried leaves, extracted with acetone and water, drug: extract ratio 35–67:1) contain 22–27% flavone glycosides and 5–7% terpene lactones, of which approximately 2.8–3.4% consists of Gingkolides A, B and C and 2.6–3.2% bilobalide. The level of Gingkolic acids is below 5 mg/kg (WHO Monographs, 1999).

#### 25.6. Quantified extracts

These are extracts having constituents with known therapeutic or pharmacological activity. Groups of compounds likely to have the desired pharmacological activity are unknown, but are not solely responsible for the clinical efficacy of the extract. The monograph must define a range of content of the selected constituent(s) some of which are lead compounds.

Standardisation by blending different batches of a herbal drug before extraction, or by mixing different lots of herbal drug preparation, is acceptable but adjustment using excipients is not acceptable (Heinrich et al., 2004).

Examples of Quantified Extract.

Gingko biloba leaves.

Gingko biloba leaves are used for improving cerebral and peripheral circulation among old people and also for cerebral circulation. Gingko extracts are well-known for the two groups of compounds which are particularly relevant:

The Flavonoids (0.5-1%): flavone and flavonol glycosides, acetylated flavonol glycosides, biflavonoids.

Terpene lactones: (0.03–0.25%).

Although it is a well-known fact that these compounds are among the most important ones, other compounds are also important and their role and action must also be understood as they may well add to the pharmacological activity of the extract.

## 25.7. Quality control

Microscopy is less important here as opposed to phytochemical methods. In the case of the crude drug, Thin Layer Chromatography is only feasible for some components like the flavonoids. The presence of vital minor compounds may be masked and may not be very visible. It is in this particular instance that the complementarity of analytical methods like HPLC and GC are of paramout importance for analyzing both the lead and the minor compounds.

#### 26. Side-effects (toxicity) of plant extracts

Botanical secondary compounds are not benign molecules; ecologically speaking, these evolved as chemical defences that can repel, stun, poison or kill other species. It would be naïve on anybody's behalf to think that every plant extract is necessarily safe for human consumption. It is precisely for these reasons that poison centers have been established across several continents.

Nonetheless, it would be difficult to distinguish between an effective medecine from a deadly poison. The dosage is critical in these circumstances especially as some plants with a long history of use have been implicated as being potentially toxic. Among such plants are, amongst others, the Comfrey (*Symphytum officinale*) which has been reported to cause acute liver damage; Yohimbe (*Corynanthe yohimbe*) is used as a dietary supplement and aphrodisiac but at high doses, it is suspected able to cause kidney failure and death.

# 27. Adulteration

Over and above the dosage factor, is adulteration either accidentally or intentionally. Foxglove (*Digitalis purpurea*) has shown up as an adulterant in herbal mixture where it is not among the listed ingredients. Sometimes herbal remedies have been found to be tainted with heavy metals.

In other instances, when one medicinal plant has been replaced by another similarly looking one, this led to disastrous consequences. Although *Aristolochia* species are used in Chinese medicine, the replacement by *Aristolochia fangchi* has caused the death of several patients who were attending a weight loss clinic in Belgium (Schaneberg and Khan, 2004).

# 28. Microbial contaminations

With requirements of Good Agricultural Practices (GAP) and Good Manufacturing Practices (GMP) being improved more and more on producers of medicinal plants, Microbial contamination can be controlled. It is a fact that natural materials harbour a large number of spores and other microorganisms but nonetheless, the maximum number or microorganisms allowed is regulated in most Pharmacopeia. The European Pharmacopeia, for example, excludes the presence of *Escherichia coli* and *Salmonella* sp. and limits aerobic microorganisms up to  $10^5$  per g or ml and this includes up to  $10^3$  yeast and fungi per g or ml and up to  $10^3$  enterobacter per g or ml (Heinrich et al., 2004).

# 29. The convention for biological diversity

It is a fact that the role of ethno-botanist is crucial in the search for new drugs and this role has become so much more significant in the second half of the 20th century. The study of ethnobotany has gained in importance and the 'Western' use of such information has come under increasing scrutiny. National and indigenous rights on these resources have become acknowledged by both academic and industrial researchers. It is also recognized that the need for basic scientific investigations of plants used in indigenous medical systems is becoming ever more relevant.

The relevance of such data coupled with the ever-increasing rights expected from communities on their data along with the battle for conservation, have re-shaped the entire approach towards bioprospecting.

The major ideas concerning bioprospecting and conservation were first presented by the American scientist Thomas Eisner (1989, 1991). He entitled the activity 'chemical prospecting' and described it as 'the exploratory process by which new, useful natural products are discovered' (Eisner, 1991). He proposes chemical prospecting to be substantially intensified. He also highlighted the loss of species and the concomitant loss of chemicals of great value for the progress of medicine.

The penning of the Convention on Biological Diversity (CBD) (UN Convention on Biodiversity) ushered a new era in natural products drug discovery and development. The CBD encompasses all of Eisner's ideas and takes into account the element of benefits to the provider (state and their people). It was opened for signature at the UN Conference on Environment and Development in Rio de Janeiro in 1992. By January 1999, 175 states and the European Union had ratified the Convention. Thus the CBD is one of the international conventions with the highest number of State Parties. It entered into force after being ratified by 30 States in December 1993. The US is one of the States not being Party to the Convention.

#### The objectives of the CBD as spelled out in Article 1

The objectives or the Convention, to be pursued in accordance with its relevant provisions, are the conservation of biological diversity, the sustainable use of its components and the fair and equitable sharing of the benefits arising out of the utilization of genetic resources, including by appropriate access to genetic resources and by appropriate transfer of appropriate technologies, taking into account all rights over those resources and to technologies, and by appropriate funding.

The CBD regulates a number of important issues related to the global conservation and sustainable use of biological resources and covers a much broader field than access to genetic resources.

In Article 15, the CBD also called for recognition of the sovereign rights of countries to control utilization of their natural resources and genetic materials while Article 8, addresses issues related to the global conservation and sustainable use of biological resources, and it covers a much broader field than access to genetic resources.

Article 15 should-as the Convention in general- be seen in a North to South perspective. Broadly speaking, it gives citizens and companies of developed countries access to the genetic resources in developing countries—on certain conditions. One them is the obligation for developed countries to provide and facilitate access for and transfer to developing countries, or technologies that make use of genetic resources. This is spelt out in Article 16.

Article 15 is in itself a compromise between these interests. In Paragraph 1, it makes a case for the legal starting point concerning sovereign rights of States over their natural resources.

"Recognising the sovereign rights of states over their natural resources, the authority to determine access to genetic resources rests with the national government and is subject to national legislation".

Countries in the North achieved acceptance of their demands for access to genetic resources from the developing countries. On the other hand, the developing countries achieved the recognition of national sovereignty over genetic resources, and the principle of prior informed consent and benefit sharing as a basic condition for access.

Broadly speaking, the developed countries had two main interests in biological diversity. The growing scientific recognition and political concern for the extinction of species and biotypes lead to a demand for stronger conservation efforts. This concern is also addressed in Article 3 of the Convention:

Article 3 of the Convention expresses the principle or state's soverign right to exploit their own resources pursuant to their own environmental policies, combined with an obligation to ensure that activities within their jurisdiction or control donot cause damage to the environment or other states or areas beyond natural jurisdiction. This principle is firmly established in international law'.

Developed countries had, as well a commercial interest in the exploitation of the genetic material through bio-prospecting; the use of genetic materials in scientific research, and as a basis for new products in pharmaceutical, medical, agricultural and other industries. Such use in the short-term perspective pre-supposes access possibilities. Access in a long-term perspective pre-supposes both effective conservation and access legislation. This feature is also embodies in the Articles of the CBD namely:

While confirming the state's soverign right over its genetic resources, the CBD Article 15 nevertheless implies a certain limitation on the exercise or this right. The State is not entirely free to decide on the use or its genetic resources but it has an '...obligation to create conditions to faciliatate access to genetic resources for environmentally sound uses by other Contracting Parties'.

The developing countries, on the other hand, had sovereignty over the resources as their main objective. This was seen as a pre-requisite for receiving benefits from the use of their biodiversity. Furthermore, such benefits would increase these countries possibility to make conservation efforts. Exploitation of genetic material for research and industrial purposes adds new economic value top the forest areas. This is also taken on board in Article 15 of the CBD.

Article 15, Paragraphs 4 and 5, imply that access to genetic resources should be granted by the source state through a system of individual permit and/or mutual agreement between the interested parties.

# 29.1. Mutually agreed terms (MAT) and prior informed consent (PIC)

As mentioned above, CBD Article 15, Paragraphs 4 and 5, contain the basic legal conditions for access to genetic resources. Access when granted shall be on 'mutually agreed terms' (MAT) and shall be subject to Prior Informed Consent (PIC) by the providing parties or 'unless otherwise determined by that Party'. MAT and PIC are general principles of the CBD and is expressed in several articles as well.

MAT and PIC have significantly changed the basic conditions for research involving traditional knowledge in particular ethnopharmacological research.

Countries and Peoples providing the resources for natural products research and drug development now have well-defined benefits and rights. This will have a direct impact on the sharing of benefits accruing from collaborations.

It goes without saying that these clauses of the CBD will and have had a definite impact on bioprospecting. This exercise, which focuses on the development of new drugs with enormous financial returns for big international companies, requires high-throughput screening systems and extracts. For this to materialize correctly, cooperation, collaboration and trust are pre-requisites between donor countries and international organizations.

It is therefore clear that the development of new drugs will benefit from the breadth of the contribution of ethnobotany and ethnopharmacology, from the indigenous and orally transmitted medical systems to drug development. Modern science stands to benefit from the conservation of plant specimens and also on the preservation of oral traditions, which have stood the test of time in some primitive societies.

# **30.** Conclusion

This paper has given an overview of the importance of medicinal plants from antiquity to date date. It cannot be denied that pharmacognosy has had a chequered history but has evolved over the years to become one or the pillars of areas like pharmacy, medicine and natural product chemistry amongst others. All these scientific disciplines now recognize the importance of plants as sources of medicines and have initiated active research programmes either to isolate new lead compounds or to produce standardized extracts.

With the estimated 10–100 million species or organisms living on earth and higher plants forming a group of some 250,000 species out of which only 6% has been investigated for biological activities and 15% for their chemical constituents, it looks increasingly like we have only scratched the surface of this world's wonderful resource.

While the pharmaceutical industry in the developed world, will continue to investigate promising leads from natural products in their effort to produce new drugs entities, the production of new medicines in the developing world may have quite different priorities. In these parts of the world, when a plant is readily available and has the potential to provide inexpensive therapy for the treatment of a disease, then a product may well be developed. Close collaboration is expected between clinicals and scientists with a common endeavour—production of safe, quality and efficacious products.

Pharmacognosy, an area or science once considered moribund, has a very bright future ahead as it will continue to provide new lead molecules for major ailments facing us. Several hard evidence has been given in the text to back this evidence and it can only be hoped that research will shed more light on these facts provided that the critical mass of researchers are involved and enough funds put to disposal.

Nonetheless, issues like conservation of both ethnobotanical data and biodiversity must addressed as ignoring it will pose a serious challenge to the search of new leads.

With the interest that has been generated both the general public, university researchers and multinationals across the globe into natural products, there is now more than ever a golden opportunity to continue making worthwhile contribution to healthcare.

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