Chapter 1

Introduction

1. INTRODUCTION

Last century has been very remarkable and unique in the entire history of medical sciences. In fundamental sciences, discovery of double helix and completion of human genome project were achievements. In diagnostics, technologies like MRI and PET scan were the achievements. In surgery, bypass, transplants and prosthetics changed the life expectancies upwards dramatically (Patwardhan, 2005). Developing countries are distinguished from developed world through three key characteristics- disproportionate population growth, disproportionate infectious diseases and health care going out of reach. A WHO data showed that density of physicians per 1, 00,000 persons in various countries as: - Rawanda1.87, Ethiopia 2.85, Uganda 4.70, Benin 5.75, India 51.25 and China 164.24. In contrast to the numbers for Australia and USA as 249.13 and 548.91 respectively, clearly shows the need of health care in developing countries (WHO, 2004). As per the World population prospects, an estimated 7.4 million people are living with HIV in China, Indonesia and Viet Nam, 5.1 million in India, 1.6 million in Latin America and 8.25 million people in Africa(World Population Prospects). There are at least 300 million acute cases of malaria each year globally, resulting in more than a million deaths. Around 90% of these deaths occur in Africa, mostly in young children. Against that, malaria is virtually nonexistent in developed world. The number of persons who suffered from all types of tuberculosis in 2002 was 523 per1, 00,000 in South Africa and South East Asia together, out of which 122 died, whereas in US and Europe the number was 97 and 14. Some 3 million people mostly women and children die every year from diarrhoeal diseases (Aldhous, 2003; Triggle, 2003). Leaders, policy makers and scholars all over the world are struggling with possible solutions to these complex problems as there is no single solution. One among the possible solutions is the place and utility of Traditional Medicines (TM) as an effective and efficient vehicle which is affordable to the large population in the developing countries. TM has become an extremely valuable commodity for the world today, because it provides what the world misses the most. The low income developing countries miss the modern medicine because they cannot afford it whereas high income developed world misses the holistic approach in TM.

1.1 Recent Trends in Herbal Medicines

Use of indigenous drugs from plant origin forms a major part of Complementary and Alternative Medicine (CAM) or Traditional Medicine (TM) (Joshi and Chavan,

2004; Theoharis, 2004).TM is attracting more and more attention within the context of health care provision and health sector reform. Many factors are contributing to widespread use of TM (Kawasar and Sayyed, 2003). TM is a comprehensive term used to refer to both to TM systems like traditional Chinese medicine, Indian Ayurveda, Arabic Unani medicines and to various forms of indigenous medicine. TM therapies involve use of herbs, animal parts and minerals. It also includes acupuncture, manual therapies and spiritual medicines. As per the report of the Inter Regional Workshop on Intellectual property Rights, the world market for TM including herbal products and the raw materials has been estimated to have an annual growth rate of 5-10%. Total global herbal market is estimated as US $ 5 trillion by 2050. The Indian medicinal plant based industry is growing at the rate of 7-15% annually. The value of medicinal plants related trade in India is estimated at Rs. 5000 crores per annum. Global trend leading to increased demand of medicinal plants for pharmaceuticals, phytochemicals, neutraceuticals, cosmetics and other products is an opportunity sector for Indian trade and commerce (Singh and Khanuja, 2003).

WHO defines TM as diverse health practices, approaches, knowledge and beliefs incorporating plant, animal and/ or mineral based medicines, spiritual therapies, manual techniques and exercises applied singularly or in combination to maintain well being as well as to treat, diagnose or prevent illness. The focus of TM, CAM, Ayurveda and other alternative systems of medicines is patient rather than disease. These systems aim to promote healthy and enhance quality of life. The concept of constitutional uniqueness of human individuals leading to prescription of suitable drugs and specific diet is a remarkable feature of TM and CAM (Patwardhan, 2006). Ayurveda was developed through daily life experiences with the mutual relationship between mankind and nature. The plant species mentioned in Ayurveda may be explored with the help of modern scientific approaches for better lead in the health care (Mukherjee and Wahile, 2006). The knowledge of herbal medicines allows drug researchers to start from a well tested and safe botanical material. Ayurveda follows a true reverse pharmacology approach by changing the course of normal drug discovery from laboratories to clinics instead from clinics to laboratories (Vaidya, 2002). The reverse pharmacology approach could save time, cost and toxicity. Safety remains most important starting point and efficacy becomes a matter of validation (Patwardhan, 1999). Globally there is a positive trend towards holistic health, integrative sciences, systems biology approaches in drug discovery and therapeutics

that has remained one of the unique features of Ayurveda. A golden triangle consisting of Ayurveda- Modern medicine- science will converge to form a real discovery engine that can result in newer, safe, cheaper and effective therapies (Mashelkar, 2003).

30% of the worldwide sales of drugs are based on natural products. Though recombinant proteins and peptides account for increasing sales rates, the superiority of low molecular mass compounds in human disease therapy remains undisputed mainly due to favorable compliance and bioavailability properties. Approaches to improve and accelerate the joint drug discovery and development process are expected to take place mainly from innovation in drug target elucidation to lead structure discovery. Therefore, the need for new concepts to generate collection of large compounds with improved structural discovery has been correctly emphasized by Garbley and Thierick (1993). The mass screening of plants in the search for new drugs is expensive and inefficient. It would be cheaper and perhaps more productive to re- examine plant remedies described in ancient and medieval texts (Holland, 1994).

TM dates back thousands of years and still widely used in Asia (Bedi, 2002; Ernst, 2000). Use of TM has recently become increasingly popular among the patients seeking alternative treatment options (Blance, 2001; Bielory, 2000). The number of visits to alternative medicine practitioners in US is growing rapidly. In 1997, the number of visits was estimated to be 629 million which exceeded the number of visits to all primary health care physicians (Neldner, 2000). Approximately US $ 27 billion was spent for alternative therapies in 1997 and US $ 3.24 billion on herbal therapy (Klepser and Klepser, 1999). Most patients are getting attracted to TM because conventional therapy has failed or they feel there are no side effects as the products are natural (Eisenburg and Kessler, 1993). WHO estimated that 80% of the world's population relies on herbs for its primary healthcare needs (Koushal and Singh, 2001).There are some traditional medical practices such as Ayurveda and Siddha in India, Chinese medicine in china, Unani in Islamic countries. More than 35,000 plant species are being used in traditional and ethnomedicinal practice. TM provides the surest means to achieve total healthcare coverage of the world population. The modern medicines are expensive, painful and with side effects whereas TM is safe and economically feasible (Katewa and Arora, 2001; Latha, 1999). The most common TM providers were massage therapist (19.4%) and herbalist (12.4%). The most common herbs were Chamomile (13.1%) and Aloe Vera (8.5%). The most common

nutritional products were Ginseng (3.6%) and Ginkgo Biloba (2.8%) (Rivera, 2002). India is the largest producer of medicinal herbs and is called as botanical garden of the world (Dubey, 2004). India enjoys the benefits of varied climate from alpine in Himalayas to tropical wet in South and arid in Rajasthan. Such climatic conditions have given rise to rich and varied flora in the Indian subcontinent. India officially recognizes over 3000 plants for their medicinal value. Over 6000 plants in India are used in traditional, folk and herbal medicines fulfilling about 75% of medicinal needs of the third world countries (Rajshekharan, 2002). The R & D thrust in the pharmaceutical sector is focused on development of new drugs, innovative/ indigenous processes for known drugs and development of plant based drugs through investigation of leads from traditional systems of medicine. Neutraceuticals and cosmeceuticals are of great importance as a reservoir of chemical diversity aimed at new drug discovery and are explored for antimicrobial, cardiovascular, immunosuppressant and anticancer drugs (Patwardhan, 2004). Natural products including plants, animals and minerals have been the basis of treatment of human diseases (Hooper, 1992; Singh, 1997; Garbley, 1999). Numerous molecules have come out of experimental base, examples include rauwolfia alkaloids for hypertension, holarrhena alkaloids in amoebiasis, guggulosterones as hypolipidemic agents, *Mucuna pruriens* for parkinsons disease, piperidine as bioavailability enhancer, buccoside in mental retention, picrosides in hepatic protection, phyllanthins as antivirals, curcumines in inflammation, withanoliodes and many other steroidal lactones and glycosides as immunomodulators (Patwardhan, 2000).

1.2 Need for systematic investigation of plants

Most of the regulatory guidelines and pharmacopoeias suggest macroscopic and microscopic evaluation and chemical profiling of the botanical materials for quality control and standardization. Macroscopic identification is based on parameters like shape, size, color, texture, surface characteristics, fracture, odor, taste which are compared with standard material. Microscopy involves comparative microscopic inspection of broken and powdered crude drugs. However these parameters are judged subjectively and substitutes and adulterants may closely resemble as genuine material. TLC and HPTLC are routinely used as valuable tools for qualitative determination of phytoconstituents. Many analytical techniques like volumetric analysis, gravimetric determinations, gas chromatography, column chromatography, HPLC and spectrophotometric methods are frequently used for quality control and

standardization. The use of chromatographic techniques and marker compounds to standardize botanical preparation has limitations because of their variable sources and chemical complexity. Many factors may affect the ultimate chemical profile of any herb. Intrinsic factors such as genetics and extrinsic factors like cultivation, harvesting, drying and storage conditions. Routine chemotaxonomical studies provide only a qualitative account of secondary metabolites. For quantitative studies, specific markers can be used that can be easily analyzed to distinguish between varieties. Such metabolites being used as markers may or may not be therapeutically active but should ideally be neutral to environmental effects (Joshi, 1999). One of the problems in standardization is selection of correct chemotype of plant to which clinical effects are attributed e.g. *Withania somnifera* have three chemotypes depending upon the presence of a class closely related to steroidal lactones like withanolides, withaferin A etc. The content of these phytoconstituents and other biologically active compounds may vary depending upon the environment, genotype, time of collection of plant material etc. Another difficulty in identification of certain plant species is that it may be known by different binomial botanical names in different regions. Eg Shankhapushpi which is an important Medhya Rasayana in Ayurveda is known as *Cansora decussate, Evolvulus alsinoides* and *Clitoria ternate* (Pandey, 2002). To overcome these problems, DNA markers play an important role. DNA markers are reliable as genetic composition is unique for each species and is not affected by age, physiological conditions and environmental factors (Warude and Chavan. 2003).

Standardization of herbal drugs is not just an analytical operation for identification and assay of active principles; rather it comprises total information and controls to necessarily guarantee consistent composition of all herbs. Indian herbal drug industry needs to ensure procurement of standardized authentic raw material free from toxic contaminants. Improving processing technologies, conducting all operations under GMP compliance and maintenance of in process quality control for manufacturing quality of herbal products also need evidence for therapeutic efficacy, safety and shelf life. Such approaches remain important in global promotion of herbal drugs (Patwardhan, 2006).

1.3 Plant Adaptogens

Plant adaptogens is a class of metabolic regulators which have been shown to increase the ability of the organism to adapt to environmental factors and to avoid damage from such factors. The definition of adaptogens is as follows (Panossian 1999).

- The adaptogenic effect is nonspecific in which the adaptogen increases resistance to a very broad spectrum of stressors of different physical, chemical and biological nature.
- An adaptogen is to have a normalizing effect, that is, it counteracts or prevents disturbances brought about by stressor.
- An adaptogen must be innocuous to have a broad range of therapeutic effect without causing any disturbance to the normal function of the organism.

Pharmacological studies revealed a basic difference between classical stimulants and adaptogens in situations of forced muscular work. Stimulants give a temporary increase in work capacity and then it is followed by a marked decrease in work capacity. Extracts and glycosides from *Acanthopanax* and *Rhodolia* showed that the level of performance after reaching its maximum is not followed by a corresponding minimum of average work capacity. In stress, there is involvement of hormones, cytokines and neuroendocrine (Tache and Rivier, 1993). Plant adaptogens reduce the damaging effects of various stressors due to alteration in the reactivity of host defense system. *Oscimum sanctum* was evaluated for its antistress and adaptogenic activity in immobilization stress and cold restraint stress in rats (Dadkar, 1987). Stress induced gastric ulceration was studied. Pretreatment with aqueous extract of *Oscimum sanctum* failed to offer any protection against gastric ulcers in immobilization stress but offered partial protection in cold restraint stress. It was observed that rise in BUN levels which was evident only in cold restraint stress was partially inhibited by pretreatment with *Oscimum sanctum*. There was significant decrease in levels of enzyme SDH in liver but not in brain. This enzyme has a role in cellular respiration and tricarboxylic acid cycle (Boyer, 1963). Pretreatment with *Oscimum sanctum* proved effective in improving the enzyme levels in both the models. Adaptogenic activity of aqueous extract of the roots of *Boerhaavia diffuasa* Linn in forced swim stress and cold stress in rats. Biochemical parameters like serum glucose, cholesterol, triglycerides, SGPT, BUN were studied. These parameters were found to be altered during stress. The extract was effective in regulating blood glucose level by homeostatic mechanism and controlling the hypothalamo- hypophyseal axis activity (Mungatiwar et al, 1997). *Tinospora cordifolia* was studied for adaptogenic activity (Patil and Patki, 1997). A polyharbal formulation containing Ashwagandha, Brahmi, Shatavari, Amla, Gokhru was evaluated in forced swim induced immobility in rats.

In Indian systems of medicine, a large number of herbal drugs have been advocated for various types of diseases or stress related disorders (Arulkuraan et al., 2007). One of the main strategies is to increase natural resistance of the body to disease or stress causing agent rather than directly neutralizing the agent itself. These agents are of plant origin which induce paraimmunity, the nonspecific immunomodulation of essentially granulocytes, macrophages, natural killer cells and complement function. Rasayana are group of nontoxic herbal drug preparations which are used to improve the general health by stimulating immunity (Kumar, 1999; Sharma, 1996). These drugs act either by stimulating nonspecific system i.e. granulocytes, macrophages, T lymphocytes and different effector substances or by suppressing resistance against infections, stress and may occur as an account of environmental or chemotherapeutic factors. Both need to be tackled in order to regulate immunological functioning. Hence, both have their own standings and search for better agents exerting these activities is becoming the field of major interest all over the world. In general, adaptogens are biological response modifiers that affect the immune response in either positive or negative fashion.

Evaluation of Indian medicinal plants has been done for adaptogenic activity. According to Dahanukar and Thatte (1997), only those Rasayanas which produce sweet (Madhur) vipaka like *Tinospora cordifolia, Emblica officinalis, Terminalia chebula and Withania somnifrera* were found to stimulate the reticulo endothelial system but those with bitter (Katu) vipaka do not produce the same results. Eg *Acorus calamus, Commiphora mukul and Picrorrhiza kurroa* .Among the dugs which stimulate, *Tinospora cordifolia* which is used for its general adaptogenic and prohost immunomodulatory activity (Jagtap and Iyer, 2004). It was reported that *Tinospora cordifolia* inhibited the production of IL-1 and TNF-α by macrophages (Dhuley, 1997). *Picrorrhiza kurroa* was found to stimulate both cell mediated and humoral immunity. *Tylophora indica, Aconitum heterophyllum and Holarrhena antidyssentrica* appeared to stimulate phagocytic function (Atal, 1986). *Picrorrhiza kurroa* leaf extract was found to stimulate the cell mediated and humoral components of the immune system as well as phagocytosis (Sharma 1994). The studies on *Withania somnifrera* indicated that it could reduce the cyclophosphamide induced toxicity and its usefulness in cancer therapy (Davis and Kuttan, 1998). The decoction of roots of *Ficus benghalensis* showed a significant increase in the percent phagocytosis and phagocytic index (Gabhe and Khan, 2006). *Adhatoda vasica and*

Lawsonia inermis were studied for their immunostimulant activity. Both the plants were found to be effective in cyclophosphamide induced myelosuppression which was indicated by increase in WBC count (Jinyverghese and Karpe, 2005). *Centella asiatica* possess strong immunostimulant activity which was comparable with market stimulant drugs (Patil, 1998). A study on napthaquinones (Plumbagin, Alkanin, Shikonnin) and cytotoxic agents such as colchicines, vincristine, cyclophosphamide exert direct cytotoxic activity when applied in high doses but at the same time exhibit an immune induced cytotoxicity when applied in low doses (Wagner, 1988). Kulkarni and Karade 1998 screened other allied napthoquinone derivative namely Lawsone from *Lawsonia alba* Linn for immunostimulant activity (kulkarni and Karand, 19998). The prevention of degranulation process by boswellic acid justifies its use as immunomodulatory effect. *Azadirachta indica* is known as 'sarvaroga nivarini' i.e. cure for all diseases possesses anti HIV, antitumor, antimalarial activity (Govindachari, 1992; Ray, 1996). A study on azadirachtin- a triterpenoid from *Azadirachta indica* on the immune response of Oreochromis mossambicus has been undertaken. It was observed that azadirachtin greatly increased primary and secondary responses to BSA compared to control (Logambal, 2000). *Asparagus recemosus* which is a well known Rasayana drug was studied for its immunomodulating properties (Goyal, 2003). It has been shown to protect the rats and mice against abdominal sepsis (Dahanukar, 1986; Thatte, 1987). Percentage mortality of *Asparagus recemosus* treated animals was found to be significantly reduced while survival rate was comparable to that of the groups treated with metranidazole and gentamycin. Dhuley (1997) has reported the revival of macrophage chemotaxis and IL-1 and tumor necrosis factor (TNF) production by oral treatment of *Asparagus recemosus* root extract in orchratoxin A treated mice (Muruganadan et al., 2000). *Chlorophytum borivilium* root extract was found to be effective immunomodulator. (Thakur, 2006). Standardized extracts of Echinacea, Cat's claw and saw palmetto was each evaluated for ability to activate macrophage and natural killer cell in vitro by Groom, 2007. The fern, Selaginela was studied for its immunomodulatory and antioxidant activities (Gayatri, 2005). A herbomineral formulation 'Suvernamalini Vasant' was evaluated for its immunomodulatory activity in *E. coli* induced peritonitis in mice and on phagocytic function of polymorphonuclea (PMN) leucocytes in rats (Sangle and Darp, 2004). Sphaeranthus indicus Linn was evaluated in cyclophosphamide induced myelosuppression, HA titre and, DTH response and

phagocytic activity. Active principles of flower heads were responsible for stimulation of cell mediated response and phagocytic function (Bafna and Mishra, 2004).

Drugs included in the Rasayana category are known to possess the adaptogenic and rejuvenating properties. But there are many plants which are not known as Rasayana but are used in the formulations which are used as tonic along with Rasayana drugs. *Pistacia integerrima* and *Hedychium spicatum* were among those drugs. These were used in formulations like Chyavaprash, Piptonil, Kumariasav, Balacatudrika curna, Sringyadi Curna, Brainokan, Cardiokan and Kantakadi avaleha along with the well known Rasayana drugs like Shatavari, Ashwagandha, Baheda, Aloe etc.

1.4 Screening methods for adaptogens

Numbers of in vitro and in vivo test systems are available for screening adaptogenic activity. For selection of proper test models, the understanding of methodologies is required which enable the researcher to thoroughly evaluate such agents. However following are few selected methods widely employed for evaluation of adaptogenic activity.

1.4.1 *E. coli* induced abdominal sepsis

The immunoprotective effect was assessed by *E. coli* induced abdominal sepsis model which was carried out in two parts. In the first part the strength of E. coli was standardize to induce 100% mortality. In the second part the effect of *E.coli* injection in mice and protection is evaluated (Thatte, 1988).

1.4.2 Carbon clearance test

This is the simplest model to test Phagocytosis. It is the test of phagocytosis efficiency and can be correlated with in vitro granulocyte test. Drug was administered to mice orally, after 24 h each mouse was given intravenous injection of 0.3 ml of Indian ink dispersion per 30g body weight. Blood samples from retro orbital plexus are taken at intervals of 3, 6, 9, 12, 15 min. The rate of elimination of carbon clearance from blood is determined by turbidometric spectroscopy at 650 nm (Biozzi et al. 1953).

1.4.3 Cyclophosphamide induced myelosuppression

Cyclophosphamide induced myelosuppression was studied according to the method described by Pallabi et al., (1998). Different hematological parameters like WBC, RBC, Hb, HCT and MCV were determined using reported methods. The treatment was continued for further 10 days and on the 25^{th} day from the day of start of

treatment; all groups received a single dose cyclophosphamide 250mg /kg orally. On the 26th day, blood was collected from retro orbital plexus of each animal and the hematological parameters were determined.

1.4.4 Anoxia tolerance test

Albino mice of either sex were divided into four groups. Stress was induced in the animals by placing individual animal in hermetic vessel of 1 L capacity. Reaction to anoxia stress was recorded as anoxia tolerance time i.e. first sign of convulsion was considered as end point. The study was conducted for 21 days and at the end point of 1st, 2nd and 3rd week anoxia stress tolerance time was recorded with all animals. (Hirumalima, 2000; Suberans, 1993)

1.4.5 Forced swim test

Stress is exerted by keeping the rats in cylindrical vessels (length 48 cm and diameter 30 cm) filled with water to a height of 25 cm over period for two hours daily for 7 days. Animals are sacrificed at the end of specific period and blood is collected for biochemical studies (Mungatiwar et al., 1997).

1.5 Selection of plants for study

While going through the literature, different plants employed in traditional systems of medicine for different disorders, some drugs are specifically mentioned for their rejuvenative properties which are commonly known as Rasayana. In our literature search, it was observed that there are number of drugs which are not included in Rasayana but still mentioned to have restorative and rejuvenative properties and used in several ayurvedic formulations like Kumari Asav, Chyavanprash, Kantakadi Avaleha etc. Some of these drugs although subjected for some stray studies, still could be explored for providing rationale between the phytochemical profile and pharmacological activity, hence generate a need to evaluate them considering this aspect.

In our efforts to screen herbal drugs, leads were taken from Ayurveda and two plants viz., *Pistacia integerrima* (leaf galls) and *Hedychium spicatum* (rhizomes) were selected for detailed phytochemical investigation and assessment of their biological activities, as they are widely used in formulations.

1.6 Review of literature on the selected plants species

Pistacia integerrima

Common (Indian) Names

Sanskrit- Karatshingi

Hindi-Kakara shingi

Telgu- Kakarashingi

Tamil-Kakatashingi

Gujrathi, Marathi- Kakadshingi

Family- Anacardiaceae

Habitat-Himalayan Mountain ranges on the North West from Kashmir to Shimla.

Useful parts- Leaf galls

Traditional uses-Galls are used in cough, asthma, fever and respiratory disorders. It is used as tonic and stimulant. Galls are used in form of decoction or lotion as gargles to suppress haemorrhage from gums. It is also used to suppress bleeding from nose. Hakims consider galls useful in pulmonary infections, diarrhea and vomiting (Nadkarni, 1976).

Preparations- The galls are used in Chavanprash, Kumari Asav, Balacatudrika curna, Sringyadi Curna and Kantakadi avaleha.

Phytochemistry and Pharmacology

The galls contain 20-75% tannins, an essential oil and 55% resin. The following compounds were isolated from petroleum ether extract of galls: two isomeric triperpenic acids designated pistacienoic acid A ($C_{30}H_{46}O_3$) and pistacienoic acid B, triterpene alcohol probably tirucallol, β sitosterol and a waxy compound. Two triterpenic acids were reported. (Wealth of India, Raw materilas) Steam distillation of galls gave essential oil in a yield of 1.3-1.8%. The oil was colorless when fresh and turned yellow on keeping. It has a characteristic odor of the drug. The oil is used as carminative in moderate dose it has an antispasmodic action on involuntary muscles. It has CNS depressant effect. The tetracyclic triterpenes, pistacigerrimones A, B, and C have been isolated. The essential oil from galls contain α pinene, β pinene, α phellandrene, β phellandrene, α & β terpineol, limonene. The oil is reported to exhibit CNS depressant activity (Wealth of India).The galls formed on the leaves by hemipteris were found to be effective in nose bleeding, cough, fever, loss of appetite and stomach disturbances (Chopra et al., 1965). Kalidhar and Sharma reported presence of eriodictoyl, leuteolin and dihydroquercetin (Kalidhar, 1985). Fisetrin,

fustin, quercetin, taxifolin and gallic acid has already been reported to be present (Tadakuzu, 1966). Jadhav and Laddha (2005) had reported presence of chebulinic acid in galls.

Hedychium spicatum

Common (Indian) Names

Sanskrit- Kapura Kachali

Hindi- Sitruti

Gujrati, Hindi, Marathi, Punjabi- Kapurkachri

Family- Zingiberaceae (Scintaminaceae)

Habitat- Found in subtropical Himalayas, growing abundantly in Punjab and Nepal.

Useful Parts- Rhizomes

Trditional uses- Rhizomes are reported to be stomachic, carminative, bitter tonic and stimulant. It is also used in cosmetic preparations to promote hair growth. It is useful in inflammation, asthma, pains, foul breath, and bronchitis. It is good in liver complaints, vomiting and diarrhea. The rootstock is laxative, tonic to brain, emmenagogue and expectorant. It is a well known antistress agent (Kirtikar, 1983).

Preparations: The rhizomes are used in Chyavanprash, Brainokan, Cardiokan, Vomin syrup and tablets, Piptonil syrup and Pipton tablets. It is used in hair oil like Madhavi hair oil.

Phytochemistry and Pharmacology

The plant is found to contain essential oil, starch, organic acid and a glucoside. The essential oil is reported to contain ethyl ester of p- methoxy cinnamic acid, d-sabinene, cineole, sesquiterpenes and pentadecane (Sharma, 1974). It has been observed that drug has significant vasodialatory effect in coronary blood vessels. The total alcoholic extract showed mild hypotensive action in higher doses. The total alcoholic extract also showed antispasmodic action on guinea pig ileum. Clinical studies conducted by Chaturvedi and Sharmaon *H. spicatum* indicated the antiasthmatic and antihistaminic effects of the drug. Sharma and Sharnma, (1975) studied the effect on cardiovascular system and showed that total alcoholic extract has an inhibitory effect on the cardiac activity. The benzene fraction showed presence of sitosterol and sitosterol-β-D glucoside. A furanoditerpene had been isolated from *H. spicatum* by sharma and Tondon (Sharma, 1976; Sharma, 1975). Albert and Garfagnoli reported presence of sesqueterpene alcohols. The hexane fraction was

found to possess anti-inflammatory activity (Srimal,1984). Recently two labdne diterpenes were reported (Reddy, 2009).

1.7 Research envisaged

Survey of literature on selected plants drugs i.e. *Pistacia integerrima* (leaf galls) and *Hedychium spicatum* (rhizomes) revealed that ,although, these form as components of formulations used as rejuvenating agents, these are not included in the Rasayana category . The scientific data is therefore unavailable as regards to their potential as rejuvenative agents hence these plants attracted our attention for a detailed investigation. The studies on these selected plants were planned taking WHO guidelines for quality control methods into consideration in the following manner:

1. Pharmacognostic studies:

- Collection and identification of plant materials
- Macroscopic evaluation
- Microscopic evaluation

2. Proximate analysis

3. Phytochemical studies:

- Preliminary phytochemical screening of successive extracts using qualitative chemical tests and TLC profiles.
- Preparation of selective extracts for biological screening.
- Fractionation of extracts to identify bioactive fraction.
- HPTLC finger print profiles of extracts and fractions.
- Isolation and characterization of phytoconstituents from fractions.

4. Biological screening:

1. Acute toxicity studies

2. In vivo screening of extracts and fractions for adaptogenic activity.

- *E. coli* induced abdominal sepsis in mice
- Cyclophosphamide induced myelosuppression in mice
- Carbon clearance test in mice
- Forced swim induced stress in rats
- Anoxia stress tolerance

5. In vitro screening of hepatoprotective activity of fractions.

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