PHYTOCHEMICAL STUDIES AND INVITRO BIOLOGICAL ACTIVITIES ON
LEUCAS ASPERA AND ACHYRANTHUS ASPERA

INTRODUCTION

The term ‘Pharmacognosy’ was coined by German scientist Seydler in 1815. The pharmacognosy was derived from Greek word *Pharmalcon* (a drug which is meant for dried herb) and gnosis (to acquire knowledge). Hence pharmacognosy, which literally means knowledge of drugs of pharmaceuticals [Kirtikar and Basu, 1918]. The earth is at present covered with 2,50,000 to 5,00,000 species of plants [Borris, 1996]. Out of them a small percentage of medicinal plants are on record [Moerman, 1996]. Hippocrates [in the last 5th century (B.C)] mentioned about 300-400 medicinal plants [Schultes, 1992]. The Indian vedas (*Rigveda*) are still older (3000 B.C) that reflects medical description. Charaka and Susruta were renowned ancient Indian physicians and Charaka classified about 300 vegetable drugs in to 50 groups [Kirtikar and Basu, 1918]. Dioscorides, in first century A.D mentioned about medicinal plants in *De Materia Medica*, which was considered as prototype for modern pharmacopoeias. The fall of ancient civilization had made destruction or loss of many old documents of medicinal plants and modern pharma had started only when Francois Megendic and Claude Bernard of 19th Century introduced experimental procedures in animals [Stockwell, 1988]. Later Moerman contribution on medicinal plants was very significant who had reported that 1625 species of plants in America were used in food industry and 2564 plants were found as drugs [Klink, 1997]. The use of medicinal plants has gained momentum with the continued search and experience of many generations of physicians and herbal practioners. The plant products are presented in 14 of the 15 therapeutic categories of pharma preparation and they form an important role of health care system in western world [Phillipson and Anderson, 1989]. 25% medicine of all prescription in U.S.A is from natural products and another 25% medicines are the modification of natural products [Franswart, 1990]. Plants volatile oil are generally isolated from plant material by distillation method and they contain terpenoids, monoterpenes (C10), sesquiterpines (C15),
diterpine (C20) and some low molecular weight aliphatic hydrocarbons, acids, alcohols, aldehydes, acyclic esters or lactones and exceptionally nitrogen and sulphur containing compounds, coumarins and homologous of phenyl propionoids [Dorman and Deans, 2000]. The secondary metabolites are potential medical procedure and applications in cosmetic, food [Dorman and Deans, 2000; Ueda et al., 1982] and pharmaceutical industries [Pelissier et al., 1994; Sapiro et al., 1994; Cai and Wu, 1996]. Therefore these secondary metabolites are proved to have limitless important chemicals that have derived biological properties. Some of the forest indigenous plants have potential antifungal, antibacterial and antiviral properties [Okwue, 1992]. The volatile oils incase of *Ocimum canus, Ocimum gratiassum, Ocimum trichodon and Ocimum urticifolium* are potentially responsible for antimicrobial action and these volatile oils have great effect on microorganisms [Janssen et al., 1989]. Further these plants are used in folk medicines to treat different diseases like upper respiratory tract infection, diarrhea, headache, ophthalmic, skin diseases, pneumonia, cough fever and conjunctivitis. Similarly *Xylopia aethiopica* largely found in West South Africa shows antimicrobial activity against the bacteria such as *Staphylococcus aureus, E.coli, Proteus mirabilis, Candida albicans* [Boakyeiyiadom et al., 1977]. Though there are plenty of antimicrobial agents available, majority of them have a narrow spectrum of action due to the emergence of new infections which are resistant to conventional drugs that alarms the researchers for the preparation of new drug to be used in combat to microorganisms [Okigbo et al., 2005]. According to W.H.O, infectious diseases are main cause of death especially those involve in skin, mucous membrane and many infections. For instance *Streptococcus pneumonia, Bacillus subtilus, Staphylococcus aureus, Micrococcus luteus* are gram positive bacteria that causes nosocomial, waterborne, food borne, airborne and skin infections [Delauney and Errni, 1965]. *Vibrio cholera, E.coli, Haemophilus influenzae, Pseudomonas aeruginosa, Klepsiella pneumoniae, Proteus vulgaris, Neisseria gonorrhoeae and Salmonella typhi* are all gram-negative bacteria. *E.coli* causes cholera by the release of enterotoxin. It also causes waterborne infections, dysentery, urinary tract, and food borne infections. The gram-negative bacterium also causes food borne infection. *Proteus vulgaris* causes pneumonia; *Haemophilus influenzae* causes respiratory disease, meningitis, bronchitis, pneumonia and empyemia; *Pseudomonas aeruginosa* causes skin and wound infections,
nosocomial infections [Brooks et al., 2001]; Salmonella typhi causes typhoid fever and Neisseria gonorrhoeae causes venereal diseases [Brooks et al., 2001; Pelczar et al., 1993]. Asperigillus niger, Asperigillus flavus, Tricoderma vibriae, Pencillium rubrum, Chaetomium globosum, Trichophyton mentagrophytes are the moulds, where as Candida albicans is a yeast cell. The mycelium form is mould and yeasts cells do not form mycelium but resemble as fungi [Jagdish chander, 2002].

Among the infectious microorganisms dermatophyte yeast species and certain bacteria are most frequent [Caceres et al., 1991]. During the last two decades incidence of candidiasis and other fungal infections have increased especially in immuno-compressed host [Diamond, 1991]. It may be due to the non-availability of suitable drugs or toxicity of available drugs like Amphotericin-B [Maddux and Barriere, 1980]. The present thesis work predominantly represents the volatile oils and their activity on microbial and vector control properties. As such a deep study on volatile oils is needed for determining the activities of the volatile oils. The volatile oils are very complex mixture of compounds, which mainly have monoterpenes and sesquiterpenes hydrocarbons with general formula \((\text{C}_5\text{H}_8)_n\) [Svoboda and Deans, 1995]. Statistical data shows that so far 1000 monoterpenes and 3000 sesquiterpenes are structured and so many new natural compounds are being identified every year, but their biological activities are known for only some. In some plants like basal, one main constituent (methyl chavicol) predominates [Svoboda et al., 1999]. Generally the action of natural compounds is the result of the combined effect of their active and inactive compounds and most of the components of volatile oils have synergistic effect.

Volatile oils could act as a chemical defense against plant pathogenic disease [Svoboda and Deans, 1992], as a result many aromatic medicinal plants do not succumb to many diseases of the plant. Further the monoterpenes and sesquiterpenes of volatile oils can acts as strong barrier to fungal infection [Carlton et al., 1992]. The volatile oils exhibit reduction in growth of microorganisms depending on volatile oils concentration and chemical composition [Fyfe and Armstrong, 1998]. Artemisin (sesquiterpenoids) and derivatives of Artemisin are used as antimalarials [Viswakarma, 1990]. Due to that antimalarial property, W.H.O has recommended to develop artemisin as a drug for cerebral malaria. The present literature till now covers the
antimicrobial properties of volatile oils. But the present thesis is further extended to vector control by application of the volatile oils as natural products. As such a brief study on mosquito (Aedes aegypti) is necessary. Aedes aegypti is a common mosquito found all over the world except in some places like Antarctica. This mosquito transmits Arbo viruses responsible for yellow fever and dengue hemorrhagic fever (DHF) all over the world [Phillip et al., 2000]. Hence the alternative conventional substance is the use of natural product from plants [Consoli and Oliverima, 1994].

**Leucas aspera** (wild.) (Labiatae): Vern: Tummi in Telugu, Chota halcusa in Hindi. Leucas aspera (wild.) is a herbaceous much branched, erect or diffuse annual herb with 30-60cm high, more or less found all over India in cultivated fields as a weed. 35 species are found in India and only one species is found in Tropical America. The plant is fragrant and commonly used as antipyretic in villages. The juice of the leaves is used as an external application for psoriasis, chronic skin eruption and painful swellings [Chopra et al., 2002]. The flowers are used for cough and cold in children. An alcoholic extract of leaves shows antibacterial activity against *Micrococcus pyogenes*, *E.coli* etc. [Wealth of India, 1985].

**Achyranthus aspera** (Amaranthaceae) a perennial stiff erect herb, 0.2. 2.0 m high is growing up to 1000 m height. Stems are square, leaves elliptic ovate or broadly rhombate, 5. 22 cm long, 2 . 5 cm broad, and pubescent. The inflorescences are 8 - 30 cm long, with many single, white or red flowers, 3 - 7 mm wide. Flowering time is in summer. The plant is widespread in the world as a weed. In the northern part of India it is known as a medicinal plant in different systems of folk medicine. Plant volatile oil is predominant in many industries, particularly in pharmacy, clinical and food preservative industries. The oil and its constituents are well documented as antimicrobial agents. The volatile oils are complex mixtures of compounds which mainly having monoterpenes, sesquiterpenes hydrocarbons with general formula (C₅H₈)n. Compounds in the seeds of A. aspera are the saponins A and B. They are glycosides of oleanolic acid. The carbohydrate components are the sugars D-glucose, L-rhamnose, Dglucuronicacid (= Saponin A). Saponin B is the β-D-galactopyranosyl ester of Saponin A. The content of free oleanolic acid in A. aspera roots is 0.54 %. From the roots ecdysterone and oleanolic acid have been isolated. In the unripe seeds saponines, oleanolic acid, amino acids and hentriacontane, a long chained
carbohydrate, have been found. In the shoots an aliphatic dihydroxyketone 36,37-dihydroxyhenpentacontan-4-on and triacontanol could be found. Two long chain compounds, isolated from the shoots, have been characterized as 27-cyclohexylheptacosan-7-ol and 16-hydroxy.26-methylheptacosan-2-on by chemical and spectral investigations. The petrol extract of the shoots produced a yellow semi-solid mass. From this a pink coloured essential oil with a pleasant odour and an aliphatic alcohol (17-pentatriacontanol) were found.