PHARMACOGNOSTIC AND PHYTOCHEMICAL STUDIES ON THE SUBSTITUTES/ADULTERANTS OF CERTAIN DRUGS USED IN INDIAN SYSTEM OF MEDICINE

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By

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Chapter 1

General Introduction

India is unique country and has one of the oldest, richest and most diverse cultural traditions associated with the use of medicinal plants from the prehistoric times. This knowledge, accessible from hundreds of medical texts and manuscripts in the form of Vedic literature as a channel through which this continuous medical tradition reached down to the earliest systematisers. Ancient Indian literatures incorporate a remarkably broad definition of medicinal plants and considers 'all' plants as potential sources of medicinal substances ("Jagatyevananaoushadham na kincit vidyate dravyam vasannaanartha yogayoh" (Ashtanga hirdaya. SU.9-10). Vedic Sarnhitas, Nighantus (lexicons) and regional commentaries, has developed a unique system for using plants. The wonderful reference and treatises on herbal cure that have been available in India talk about the work of Dhanwantari, Nagarjun and Charak. In India, there is systematized use of herbal drugs and has been practiced from almost the very beginning of civilization. There are at least 5 different systems of medicine practised in India such as **Ayurveda, Unani, Siddha, Homeopathy, Naturopathy and Sowa-Rigpa.**These systems contain a large number of plants with proven or highly reliable medicinal properties. India alone boasts of more than 2000 medicinal plants, while the total number of medicinal herbs in the world used in varying systems is about 70,000.

Ayurveda

Ayurveda, one of the most Ancient systems of medicine practised in India, traces its origin to Brahma, the creator of the Universe who taught the science to Prajapati, he to Ashwini Devtas and they in their turn to Atreya etc. It dates back to Vedic age, (1500-800B.C).The term comes from the Sanskrit root, Ayu (life) and veda (Knowledge). As the name indicates, it is a system which covers the entire gamut of happy human life, involving the physical, metaphysical and the spiritual aspects. Sasruta considered the major aim of Ayurveda is to help people maintain health. The treating of the disease is considered secondary.The concept of Ayurveda is as old as the "Vedas". The Rigveda, which is written between 4500-1600BC is one

of the oldest repositories of human knowledge, mentions the use of 67 plants for therapeutic use. The Yajurveda enlists 81 plants whereas the Atharvaveda is written during 1200 BC describes 290 plants of medicinal value. The next landmark was when Sasruta samhita (600 BC) recorded 395 medicinal plants.

According to Ayurveda, all the objects in the universe including the human body, are composed of five basic elements ("panchamahabhutas") namely, earth, water, fire, air, and vacuum (ether). These elements in different proportions are in a balanced state to suit the needs requirements of different structures and function of the body matrix and its parts. These five elements combine to form the three basic forces, "tridoshas", which exist in everything in the Universe and influence all mental and physical processes. From earth and air, the air principle Vata is created. Fire and water yield the fire principle pitta ; and earth and water produce the water principle kapha. All of us are born with a particular balance of these doshas. The proportions are largely determined by the balance of doshas in our parents at the time of conception. Our body type and temperament and susceptibility to illness are largely governed by the predominant doshas.

Ayurveda, by and large, is an experience with nature. Unlike in Western Medicine many of the concepts elude scientific explanation in the modern sense of term. Western Medicine relies heavily on principles of basic sciences to explain various aspects of health science (Majumdar, 1989). The concept of science is an anathema to men of Ayurveda.

The concept of science cannot be limited to physical science alone; instead it should explore physical, living and conscious phenomena. There were attempts to equate the three doshas with biochemical neurohumours liberated by brain and its nerve endings (Udupa, 1983). Thus vata is equated with acetylcholine liberated by cerebral cortex and peripheral and parasympathetic nerve endings; pitta with catecholamines liberated by the nerve endings, hypothalamus, sympathetic nerve endings and adrenal medulla and kapha with histamine secreted by brain stem.

The drugs, when administered, act by promoting or destroying the respective neurohumours or their precursors. It is also observed that a person of vata-prakriti is lean and thin with an excess of acetylcholine, that of pitta-prakriti is muscular with a predominance of catecholamines and kapha-prakriti has a heavy body with an excess of histamine.

Udupa, unlike many others who equate agni and pitta, also holds that they are

different and agni refers to hormones and equates jatharagni with hormones with intestinal secretions, bhutagni with hormones regulating liver activities and dhatragini with hormones produced by endocrine gland that regulate cellular metabolism. Such experimental studies in Ayurveda, however lead us to the trodden path of Western medicine, leaving the much acclaimed "holistic approach" to total neglect (Laping 1984; Singh and Singh, 1990).

The concept of drug in ayurveda is somewhat different from those of the western systems of medicine. The term drug derived from the French word 'drogue' (a dry herb) and is defined as "any substance or product used to modify or explore physiological systems or pathological states for the benefit of recipient". Bhesaja or ausadha is the ayurvedic equivalent of the drug. It overcomes bhesam or osa, diseases or even fear of diseases, and includes anything, material or means, used for this purpose. Therefore even food, fasting, penance, incantations, sleep, sunlight, shade and faith in physicians are prescribed in ayurvedic therapeutic for recuperation from illness.

Siddha

The Siddha system is one of the oldest systems of medicine in India. The term "Siddha" means achievement and "siddhars" were saintly figures who achieved results in medicine through the practice of yoga. In Siddha systems, the literature is in Tamil and they are practised in the Tamil speaking parts of India. The system is also called the Agasthya system after its famous exponent, sage Agasthya.

The Siddha system is largely therapeutic in nature. The principles and doctrines of this system, both fundamental and applied, have a close similarity to Ayurveda; with specialisation in latro-chemistry. According to this system, the human body is the replica of the universe and so are the foods and drugs, irrespective of their origin. Like Ayurveda, this system believes that all object in the Universe, including the human body, are composed of five basic elements such as earth, water, fire, air and sky. The food that the human body takes and the drugs it uses are all made of these five elements. The proportion of the elements present in drugs vary and their preponderance is responsible for certain action and therapeutic results.

According to tradition, it was Shiva who unfolded the knowledge of siddha system of medicine to his concert, Parvati, who handed it down Nandideva and he

then to siddhars. Thus it is called “Saiva sampradayam” (tradition of Shiv) or “Siddha sampradayam”.

Unani

The Unani system of medicine owes it origin to Greece and has a long and impressive record in India.It was introduced in India by the Arabs and Persians sometime around the eleventh century The theoretical framework of Unani medicine is based on the teachings of Hippocrates.It aims at restoring the equilibrium of various elements and faculties of the human body. The Unani system lays great emphasis on the maintenance of proper ecological balance on one hand, and on keeping water, food and air free from pollution on the other. In this process they made extensive use of the science of Physics, Chemistry, Botany, Anatomy, Physiology, Pathology, Therapeutics and Surgery. Treatment in the Unani system of medicine is done mainly through diet control for a simple disease in the initial stages followed by the administration of a single drug, failing which a compound preparation may be administered.

Yoga and Naturopathy

Yoga, which is rooted in Hindu religious principles, had been in practice for the last 500 yrs. Derived from the Sanskrit word, *yoga* meaning “union” encompasses a variety of disciplines designed to ultimately bring its practitioners close to God. According to Maharishi Patanjali, Yoga is the suppression of modifications of the mind. It offers a significant variety of proven health benefits though it is not a cure for any medical ailment. A yoga exercise increases the efficiency of heart and slows the respiratory rate, improves fitness, lowers blood pressure, promotes relaxation and reduces stress and anxiety. It also serves to improve coordination, posture, flexibility, and range of motion, concentration, sleep and digestion. It is a supplementary therapy for conditions such as cancer, diabetes, asthma, migraine and aids. It also helps to combat addictions like smoking.

Naturopathy is a system of man building in harmony with the constructive principles of Nature on physical, mental, moral and spiritual planes of living. It has great health promotive, disease preventive and curative as well as restorative potential.According to the manifesto of British Naturopathic Association,

"Naturopathy is a system of treatment which recognises the existence of the vital curative force within the body." It therefore, advocates aiding human system to remove the cause of disease i.e. toxins by expelling the unwanted and unused matters from human body for curing diseases (http://indianmedicine.nic.in).

Homeopathy

This practice originated in Germany. The word "homeopathy" is derived from the Greek words, *homios* means "similar" and *pathos* means "suffering". Homeopathy means treating diseases with remedies prescribed in minute closes, which are capable of producing symptoms similar to the disease, when taken by healthy people. It is based on the natural law of healing- "Similia Similibus Curantur" which means "likes are cured by likes". It was given a scientific basis by Dr. Samuel Hahnemann (1755-1843) in the early 19th century. Homoeopathy today is a rapidly growing system and is being practiced almost all over the world. In India it has become a household name due the safety of its pills and gentleness of its cure.This is practiced by about 10% of Indian population. This system is different from other systems of medicine in that very dilute solutions of alcoholic extracts of plants have been used and the potency is increased by dilution. The scientific principle behind this is not well known.

Sowa-Rigpa

"*Sowa-Rigpa*" commonly known as Amchi system of medicine is one of the oldest, Living and well documented medical tradition of the world. It has been popularly practice in Tibet, Magnolia, Bhutan, some parts of China, Nepal, Himalayan regions of India and few parts of former Soviet Union etc. There are various schools of thought about the origin of this medical tradition, some scholars believes that it is originated from India, some says China and others consider it to be originated from Tibet itself. The majority of theory and practice of *Sowa-Rigpa* is similar to "*Ayurveda*" . The first *Ayurvedic* influence came to Tibet during 3rd century AD but it became popular only after 7th centuries with the approach of Buddhism to Tibet. There after this trend of exportation of Indian medical literature, along with Buddhism and other Indian art and sciences were continued till early 19th century. India being the birth place of Buddha and Buddhism has always been favorite place for learning Buddhist art and culture for Tibetan students; lots of Indian scholars were

also invited to Tibet for prorogation of Buddhism and other Indian art and sciences. This long association with India had resulted in translation and preservation of thousands of Indian literature on various subjects like religion, sciences, arts, culture and language etc. in Tibetan language. Out of these around twenty-five text related to medicine are also preserved in both canonical and non-canonical forms of Tibetan literatures. Many of these knowledge were further enriched in Tibet with the knowledge and skills of neighboring countries and their own ethnic knowledge. "*Sowa-Rigpa*" (Science of healing) is one of the classic examples of it. *Gyud-Zi* (four tantra) the fundamental text book of this medicine was first translated from India and enriched in Tibet with its own folklore and other medical tradition like Chinese and Persian etc. The impact of *Sowa-Rigpa* along with Buddhism and other Tibetan art and sciences were spread in neighboring Himalayan regions. In India this system has been practiced in Sikkim, Arunachal Pardesh, Dargeling (West Bangal), Lahoul & Spiti (Himanchal Pardesh) and Ladakh region of Jammu& Kashmir etc. (http://indianmedicine.nic.in).

All these traditional systems suffered a severe setback during the British rule in India and faced almost complete neglect for about two centuries. The allopathic system was introduced and gained ground. The withdrawal of State Patronage could not harm much as the masses reposed faith in traditional systems and it continued to be practiced. After independence the Indian systems of medicine received a fresh boost under the patronage of the National Government and its people.

Government of India took several steps for the all round development of these systems. It passed laws to regulate and promote its education and training. It established research institutions, testing laboratories and standardized regulations for the production of drugs and for its practice. Department of Indian Systems of Medicine and Homoeopathy (ISM&H) was created in March,1995 and re-named as Department of Ayurveda, Yoga & Naturopathy, Unani, Siddha and Homoeopathy (AYUSH) in November, 2003 with a view to providing focused attention to development of Education & Research in Ayurveda, Yoga & Naturopathy, Unani, Siddha and Homoeopathy systems. The Department continued to lay emphasis on upgradation of AYUSH educational standards, quality control and standardization of drugs, improving the availability of medicinal plant material, research and development and awareness generation about the efficacy of the systems domestically and internationally (http://indianmedicine.nic.in).

Protection of India's Traditional Knowledge

To protect India's Traditional Knowledge and intellectual property, Council of Scientific and Industrial Research (CSIR) and Department of Ayurveda, Yoga & Naturopathy, Unani, Siddha and Homoeopathy (AYUSH), Ministry of Health and Family Welfare, Govt. Of India is developing a collaborative database (Digital Library) known as '**Traditional Knowledge Digital Library**' (TKDL).TKDL involves documentation of the knowledge available in public domain on traditional knowledge from the existing literature related to Ayurveda, Unani and Siddha indigitized format, in five international languages which are English, French, German,Spanish and Japanese. So far, the TKDL includes about 2.12 lakh medicinal formulations (Ayurveda: 82,900; Unani: 1,15,300; Siddha: 12,950), from 148 books available in public domain. Creation of TKDL – Yoga is under process and till date about 900 no. of Yoga postures from 14 old yoga books in public domain have been transcribed, which will also be video graphed and added to TKDLdatabase.

The genesis of the maiden Indian effort dates back to the year 2000, when an interdisciplinary Task Force of experts was set up by Department of AYUSH and CSIR, to devise a mechanism on protection of India's Traditional Knowledge, after the wrong patents granted on the wound healing properties of turmeric (US patent No. 5,401,504) by USPTO and on the bio-pesticidal property of Neem (EPO patent No. 436257) by EPO, came to notice which were later fought and got revoked. Pursuant to this, studies were carried out to find out the extent of misappropriation of India's traditional knowledge which show that there is a continuous rise in misappropriation of traditional medicinal knowledge of India at the International Patent Offices. The reason for this misappropriation at International Patent Offices, as identified, is that the traditional medicinal knowledge exists in local languages, such as Sanskrit, Urdu, Arabic, Persian and Tamil which is neither available nor can be understood by patent examiners even in case of availability, at international patent offices since the information exists in local languages. In other words, there exists a language and format barrier due to which patents are being taken on the existing traditional knowledge of India. To break this language and format barrier Traditional Knowledge Digital Library (TKDL) was created by India, which with the help of Information Technology tools and a novel classification system i.e., Traditional Knowledge

Resource Classification (TKRC), makes the knowledge available to patent examiners in patent application format and in a language that can be understood by them. TKDL has enabled inclusion of approx. 200 subgroups instead of few subgroups available earlier in the International Patent Classification (IPC). The decision was taken in 35th IPC Union Meeting of World Intellectual Property Organization (WIPO) on the initiative of India. This is likely to have significant impact on the system of search and examination while granting patents in the area of traditional knowledge whereby the possibilities of grant of wrong TK patents shall get significantly reduced (http://indianmedicine.nic.in).

Herbal Medicines

In addition to the above systems wherein a large number of plants are used for alleviating the suffering of people, a large number of plants also are being used in remote areas of India and other countries. These plants are being "rediscovered" by the ethnobotanical surveys and are being used as medicines as such. The studies on bioprespecting and chemoprospecting also unearth a good number of plants having pharmacological activity. These "new" plants discovered recently add to the "herbal medicines" practised all over the world.

Active Principles

There is an ever-growing demand of medicinal plants in the global market. To approach the Western Market, at times, we have to equate our knowledge in terms of the Western Medicine. Anybody in the west, who is interested in plant medicines, would wish to satisfy himself with the data on active principles, shelf life, the pharmacological activities and side effects if any. Therefore we have to provide such data to all the customers for a better marketing of herbs.

Every medicine owes its activity to a single or a group of pharmacological active compounds (pharmacophores). In many cases, the active principle is the major compound that occurs in appreciable quantities, but in some other cases, the major compound need not be the best pharmacophore. The famous *Vinca (Catharanthus)* alkaloids never exhibited any antineoplastic property. It was only when the alkaloid extract was fractionated, and each individual alkaloid was subjected to chemical studies, the anticancerous properties of vincristine and vinblastine (which occur in

very low concentrations in roots) were discovered. The major alkaloids in *Catharanthus* are ajmalicine and serpentine and the antineoplastic alkaloids from a very minor fraction of the total alkaloids.The existing data a number of medicinal plants only indicate that the aqueous/lipid extracts exhibit various pharmacological actions. But in many cases, the different components of a plant drug are found to exert widely different properties. In the case of Punarnava (*Boerhaavia diffusa*), the roots are found to contain punarnavoside, rotenoids such as the boeravinones A, B, C, D & E, Lignans such as liriodendrin and syringaresinol, flavones, sterols, boeravine (an isofuroxanthone) and hypoxanthine-9-L arabinofuranoside. Pharmacological studies proved that punarnavoside is the antifibrinolytic agent. Liriodendrin and hypoxanthine-9-L arabinofuranoside are found to be antihypertensive and the former is a Ca++ channel antagonist but the whole plant extract exhibits anti-inflammatory, diuretic and hepatoprotective activities probably due to the other compounds present in the plant. This clearly proves that the compounds other than the so-called active principle also are active in the healing processes attributed to the drug. Similarly in *Withania somnifera*, the total alkaloidal fraction exhibits hypotensive, bradicardic and respiratory stimulant activities, while the major group of components withanolides posses antiarthritic, immunosuppressive, antitumour and antibacterial properties.

The minor components, which are normally phenolics, are never attributed with any activity. It is in this context, Duke's (1997) observations are interesting and informative. Duke (1997) describes ferulic acid, gentisic acid, kaempferol glycosides and salicylic acid as pain relievers while ascorbic acid, cinnamic acid, coumarin, myricetin, quercetin and resveratrol are explained to be anti-inflammatory. Even the variety of chemicals and their richness (concentration) in a medicinal herb is of great value in assessing its property. Duke's database states that both coriander and liquorice contain 20 chemicals with antibacterial action; oregano and rosemary have 19; ginger17; nutmeg15; cinnamon and cumin 11; Black pepper 19; Bay 10 and garlic 13. Quantity wise, liquorice contains up to 33%; bactericidal compounds (dry weight basis), thyme 21%, oregano 88%, rosemary 4-8%, coriander 22% and fennel 1.5%.

It is the alkaloids, steroids, tannins which were considered the conventional active principle of a plant drug, but this concept is changing these days. The major compounds need not be the active compounds. A number of new compounds having pharmacological action are reported recently as also new properties are discovered for

some of the known compounds. e.g. polysaccharides and lectins are found to exhibit distinct pharmacological properties.

Antioxidants

In addition to the active principles, a plant extract, prepared either in water or alcohol contains a large number of antioxidants. According to the present knowledge, in addition to the traditional role of protecting the fats, proteins and carbohydrate, the biological antioxidants manage repair system such as iron transport protein (transfferin, ferritin, caeruloplasmin etc), antioxidant enzymes and factors affecting signal transduction, vascular homeostasis and gene expression (Frankel and Mayer, 2000). But environmental pollutants such as air/water contaminants, radiation, pesticides etc, produce a large influx of free radicals in the body and tip the balance between prooxidant (free radicals) and anti free radicals (antioxidants) in favour of the former, resulting in a cumulative damage of protein, lipid, DNA, carbohydrates and membranes leading to oxidative stress. The oxidative stress, in which the free radicals outweigh antioxidants in number, is suggested to be the cause of ageing and other diseases like atherosclerosis, stroke, diabetes, cancer and neurodegenerative diseases such as Alzheimer's disease and Parkinsonism.We normally receive a good amount of antioxidants through our diet consisting of spices, vegetables, pulses and cereals, which contain a large variety of these compounds, and their role in preventing human diseases includes cancer, atherosclerosis, stroke, rheumatoid arthritis, neurodegeneration and diabetes (Fang *et al*. 2002). At the time of oxidative stress, a good amount of external antioxidants are to be pumped into and this is the basis of various antioxidant-based therapeutics.

Nutraceuticals

Most of the herbal drugs are not marketed as drugs in western countries, but are used as food supplements having pharmacological activities. They are known as nutraceuticals. Nutraceuticals are food supplemented with compounds having pharmaceutical properties. They contain a sizable amount of nutrients in the form of carbohydrates or proteins (the major compounds) and a good dose of compounds having varied pharmacological properties. In developed countries the food, especially those which are canned are fortified, are with a good dose of minerals, vitamins etc

and are prepared to take good care of health. Calcium enriched beverages, food bars, cereals, yoghurt and fermented foods dominate the nutraceutical market in Japan; while in Europe it is omega 3 fatty acids along with calcium. A number of nutritional factors are added to nutraceuticals. They are L-lipoid factor (an antioxidant to scavenge free radicals), creatinine monohydrate (to increase muscle strength), inulin (to increase calcium absorption), L- carnitine (as a fat burner) and phytosterols (to reduce cholesterol). Recently herbal medicines in the form of plant powder or extracts (containing active components) are added to prepare nutraceuticals. For example fenugreek is added to prevent and treat diabetes or it is the isoflavones (to prevent breast cancer and to reduce the incidence of osteoporosis) added.

India can produce a wide variety of nutraceuticals. They can be prepared with all the "rasayana"drugs. The plants like Ashwagandha, Amrut, Punarnava, Shataveri, Amla etc are general tonics which can be mixed with food materials. They will take care of all body functions and regulate all body systems. These provide principles active in normalizing the metabolism and contain, in addition, a number of vitamins, minerals, co-factors and anti-oxidants. The well-known preparations like "Chavanprash, Dhanvantari, rasayana, Ashwagandhadi" etc. contain plant drugs with energy rich ghee, sugar etc. These preparations can be recommended as nutraceuticals. Global market for nutraceuticals is 40-50 billion US Dollars and this market is estimated to grow at the rate of 6% per annum. Nutraceuticals also are subjected to the regulatory systems applied to pharmaceuticals or foods. "Prevention is better than cure" is the principle of nutraceuticals.

Synergism

A plant metabolome (all the compounds present within a plant) contains about 20000 compounds. A plant extract (water or alcohol) would contain at least 200 - 2000 compounds which include all types of compounds like carbohydrates, amino acids, phenolics, vitamins etc. These compounds, if possessing any pharmacological action, will also contribute to the action of active principle within the drug. The phenolic antioxidants when present, may give a free-radical free environment or protect the active principle from deterioration. The medicinal properties of simple common phenols like ferulic acid, caffeic acid, kaempferol etc. are already explained. The beneficial activities of the compounds other than the "active principles" will be

an added benefit to the patient. In certain cases the different compounds present within are found to support various systems, which are complimentary to each other. For example in *Hypericum perforatum* (St. John's Wort) where in a number of different molecules are found to have the same activity is interesting. The plant is used for treating anxiety, depression and sleep disorders. The compounds present in plant are lipid soluble hypericins (up to 0.75%), flavonoids (2-4%), xanthones (0.0004%), procyanidins (8%), hyperforin (2-8%) and volatile oil (0.1-1%) of which hypericins, xanthones and hyperforin are characteristic to St. John's Wort. It was shown earlier that hypericin inhibits mono amine oxidase (MAO) and this increases the amount of neurotransmitters in the synapse between neurons and leads to enhanced mood (Suzuki *et al*.,1984). Recent studies have shown that flavonoids and xanthones in *Hypericum* extracts inhibit catechol-O- methyl transferase, another enzyme that catabolises neurotransmitters (Thiede *et al*, 1994). Perovic and Muller, (1995) have shown that *Hypericum* extract decreases the uptake of the neurotransmitter, serotonin, in all rat synaptosomes. Thus it appears that a number of mechanisms act synergistically to increase the neurotransmitter signal.

Adultration/substitution in herbal drugs

In the historic times the question of procuring drugs and controlling their quality did not arise as the physician himself used to take care of these aspects. He were well acquainted with the herbs used by them as medicine for treatment of an ailment. They were not only used to collect medicinal plants from the nearly forests, but also preparing various drug formulations themselves as per needs of the patients. Therefore, there was a least chance of using material other than the genuine one as medicine. But present day scenario is entirely different and has gone dramatic change during the last century on account of the shift of manufacturing process from home scale produce to industrial production and most of the herbal practitioners and plant based Pharmaceutical Companies largely depend upon the crude drug dealers and traders to meet their requirement of raw materials. Thus the supply of drugs became a booming business.

Adulteration is the substitution of the original crude drug partially or fully with other substances which is either free from or inferior in therapeutic and chemical properties. The term adulteration covers a number of conditions which may be

intentional or accidental. It often occurs when a drug is difficult to obtain or when its price is comparatively high. The adulterator chooses a exhausted drugs or apparently similar material that is cheap and readily available. Other methods of use for adulteration are addition of synthetic principles to fortify inferior products, large amounts of parts of plant other than that which constitutes the drug and worthless heavy material such as sand, stone. Since flowers form the key tool for identification of a plant, in their absence, the vegetative parts are considered for identification purpose. Similar looking leaves can mislead a person and thus cause wrong identification of the plant. Collection of the wrong plant erroneously by unskilled collectors also is a major reason contributing to the adulteration of the plant of interest. The adulteration may be unintentional due to confusion in vernacular names between indigenous systems of medicine and local dialects, lack of knowledge about the authentic plant, similarity in morphology and or aroma.

The usage of such adulterated or spurious raw materials for manufacturing medicine in place of genuine one is responsible for lowering quality and efficacy of the drug because the adulterated material may not have the active compound. Moreover, it may have toxic compound that may cause deleterious effect on human health. Consequently, reliability of the finished products at national and international level is affected and betraying the faith of people on Indian Systems of Medicine.

Quality control

Herbal medicines or herbal market suffers greatly because of the poor quality of the medicine or the inconsistency of the medicinal preparations available to the society so it's a big challenges of meeting global requirements of Quality efficacy, safety and standardization. Many factors contribute to this factor and they are the following.

a) Wrong identification of the source material.

b) Plant material collected at wrong times i.e. collected at times when the active components are not at maximum like very young plant parts, old plant parts etc.

c) Poor storage conditions which will lead to microbial contamination and aflatoxins.

d) Improper extraction methods and

e) Poor knowledge on the shelf life of the extracts etc.

All these factors lead to the poor acceptance of the herbal preparations. Therefore, practices of GAP (good agricultural practice), GLP (good laboratory practices etc.) are brought in by regulatory authorities. However the quality of medicinal preparation can be judged by finding out the amount of biomarkers or active principle in a preparation. So this makes the knowledge of Biomarkers mandatory. But Biomarkers of a large number of plants are poorly known and this contribute greatly to the poor quality of medicines available in the market.

Biomarkers

Biomarkers are the compound/s or the cells/tissue/cellular contents specific to a particular plant. They indicate the presence and availability of the plant drug in a medicinal preparation. Knowledge of the quality and quantity of biomarkers in a sample, raw material, extract or formulation is a prerequisite for marketing the products. It is also useful in judging the amount of active components in a sample and also on the genuineness of the drug. The absence of a biomarker indicates that the drug is completely adulterated. Lesser quantities of biomarkers indicate poor quality of the raw material or the sample is adulterated. Compounds other than the biomarkers indicate that the drug contains some other plant/drug.

There are basically two types of biomarkers.

1. Chemical Biomarkers.
2. Pharmacognostic Biomarkers.

Chemical biomarkers

A chemical marker is a compound, whether it is a primary or secondary metabolite, or an assortment of compounds, which are seen in plant in detectable concentrations. An ideal Biomarker should be stable, easy to isolate, characteristic and should be immune to ecological changes. Any compound occurring in appreciable concentration can be a reliable biomarker. In cases where two or more plants/samples possess the same compound, a second compound present in any one of the two in combination with the first form the Biomarker. In such cases the two biomarkers in a plant need not be of the same chemical class. They can be widely different in their properties.

Pharmacognostic biomarkers

These are the tissues, cells or cell inclusions characteristic to a particular drug plant. They are of immense use in finding out the purity and genuiness of raw material in the whole form or powder form.

Pharmacognostic markers can point on the identity of the plant. For example:

(a) The presence of aerenchyma indicates that the sample is an aquatic species.

(b) Large number of starch grains is an indication that the drug is a storage organ like the root, seed, bark etc.

(c) Bark cells denote that the source is a stem or root.

(d) The presence of palisade and stomata in large number indicates that the drug is a leaf. The location of bark cells, more vessels and tracheids, indicate that this drug is adulterated.

Export potential of herbal and medicinal plants of India

In India nearly two thousand species are reported to be of medicinal value and at present about fifteen hundred drug yielding plants are well identified and are used as medicaments. Over six thousand pharmacies are reported to be functioning in the country and there are 4,246 registered herbal medicines, and the total annual demand is of thousands of quintals of herbal drugs and the acceptance and recognition of herbal medicine is increasing day by day. The international demand for herbal drugs also has increased rapidly because Ayurvedic/herbal healthcare products are considered safe under the impression that they are derived from natural products.

Worldwide, alternative medicine is becoming popular and herbal medicine has become one of the most common forms of alternative therapy. The international herbal market is approximately $61 billion. Annual sales of herbal medicinal products (HMPs) are approximately $3 billion in Germany and $1.5 billion in the US (Smet 2002). Annual turnover of Indian Ayurvedic industry is $ 0.8 billion (Rs 35,000 million) (Anon.2001).The Indian market is growing at 15-20% per annum (Rs 7,000 million or $150 million). With world demand growing at 1% annually ($ 610 million), the size of export market for medicinal plants appears bigger than the Indian domestic market. As compared to China, which boasts of herbal exports of $ 3 billion, Indian exports are dismal - $ 100 million (Bhatt).

China has been successful in acquiring the single largest share in this export

market because of its well-designed national policy on the traditional Chinese medicine. Despite contradictory claims regarding India's share in the world market of medicinal plants, one thing is very clear that ideally it should have the second largest share, but the country lags far behind China owing to its unorganised trade system and inadequate policy. The Government of India has however been quite active since 2000 to overcome this problem, and has adopted many measures to give a boost to the export of medicinal plants. A National Medicinal Plant Board has been constituted to facilitate the conservation, propagation and marketing of important medicinal plants, and its state level counter parts are also operating to implement this mandate at the level of individual states. So far the value of export is concerned, both India and China have the same problem that most of their export is in the form of low-value added products which lowers the price (Rath,2005). ISM is also facing a problems of adulteration and other malpractices and obtaining authentic drug material as a result faith in herbal drugs has declined and this remains the greatest drawback in promotion of herbal products locally or internationally. Fail in meeting global requirements of Quality, Efficacy, Safety and Standardization is also one of the major concern for India to lesser acceptance of its herbal products internationally.

Present study

From the above discussion it becomes mandatory that all our plants/ drugs are to be subjected to a detailed and thorough study on their pharmacognostic characters, biomarkers, synergistic compounds, chemical spectrum, ash and extractive values, HPTLC fingerprinting etc. essential to maintain high quality of final products so that they are accepted to the international as well as local market, so an attempt is made here to conduct a systematic study on substitutes/adulterants of five drugs commonly used in Indian system of medicine which would help in distinguishing substitutes/adulterants from genuine drugs.

Drugs selected

(I) ***Fumaria parviflora*** **Lam. (Fumariaceae).** In local market the plant is sold under the name of `Parpata' or Pitpapra'. The whole plant is medicinal and used in all types of fevers. The plant is bitter , cooling, expectorant; constipating; increases `vata', removes indigestion, biliousness, fever, burning of the body, tired feeling, wandering

of the mind, intoxication, urinary discharges, vomiting, enriches the blood; good in leprosy.

Substitutes/Adulterants of *Fumaria parviflora*:

- ***Justicia procumbens* L. (Acanthaceae).** Part used : Whole Plant.
- ***Oldenlandia corymbosa* L. (Rubiaceae).** Part used : Whole Plant.
- ***Peristrophe bicalyculata* Nees. (Acanthaceae).** Part used : Whole Plant.
- ***Polycarpea corymbosa* (L.) Lam.(Caryophyllaceae).**Part used : Whole Plant.
- ***Rungia repens* (L.) Nees.(Acanthaceae).** Part used : Whole Plant.

(II) ***Glycyrrhiza glabra* L. (Fabaceae)** popularly known as `Liquorice'(English) and `Mullethi' (Hindi) or `Yastimadhu' or `Jethirnadha' (Gujarati). The dried rhizomes and roots of this plant is used as expectorants, anti-inflammatory, demulcents and for bronchitis. It is cooling, heavy of digestion, sweet, good for the eye, improves bodily strength and complexion, is very demulcent, improves or stimulates production of semen beneficial for the hair, improves voice, overcomes vitiation of pitta (certain enzymes or enzymatic secretions which increase metabolic and other changes) of vata (corresponding to nervous and allied factors) and of blood, inflammation, cases of poisoning, vomiting, thirst, weakuess, and wasting diseases. According to Nighanturatnakara it is also beneficial in ulcer, oedema, (s'otham) combined pathological conditions of nervous and allied factors and of blood (vataraktam), haemorrhage resulting from mutual vitiation of pitta and of blood, and fresh wound (sadyovranam).

Substitutes/Adulterants of *Glycyrrhiza glabra*:

- ***Abrus precatorius* L. (Fabaceae).** Part used : Root.
- ***Alysicarpus longifolius* (Spreng.)W. & A. (Fabaceae).** Part used : Root.
- ***Taverniera cuneifolia* (Roth) Ali. (Fabaceae).** Part used : Root.
- ***Maerua arenaria* (DC.) Hook. f. &Thoms.(Capparaceae).**Part used : Root.

(III) ***Bergenia ligulata* (Wall) Engl. (Saxifragaceae).** The rhizome of this plant is the main part or source of drug and commonly called as 'Pashanabhed'. It has been in use in indigenous system of medicine since the historic period of Charaka. As the name implies, it is considered a specific remedy against kidney and bladder stones. It has been known to have various pharmacological activities and thus has several traditional uses.This is one of the twenty one drugs that constitute the *Virataradi* or

Vellantaradi gana of Vagbhata which eradicates diseases due to *vata*, vesical calculus, gravels, dysuria, and anuria (Mooss, 1980). It is light, cool, bitter, have useful effect in cough and cold (Harsoliya *at.el*.,2011). The drug enters into the composition of preparations like *Pashanabhedadi kavatha, Pashanbhedadaya ghrita, Pushayanug churna, Putikaranjasavam, Traikantada ghrtam, Valiya Marmagulika*, etc.

Substitutes/Adulterants of *Bergenia ligulata*:

- ***Aerua lanata* (L) Juss. (Amaranthaceae).** Part used : Root.
- ***Ammannia baccifera* L. (Lythraceae).** Part used : Root.
- ***Celosia argentea* L. (Amaranthaceae).** Part used : Root.
- ***Coleus amboinicus* Lour. (Amaranthaceae).** Part used : Root.
- ***Glossocardia linearifolia* (L.f.) DC. (Asteraceae).**Part used : Root.

(IV) *Polygala senega* L.(Polygalaceae).The root of this plant is popularly known as snakeroot and has sustained a reputation in the past, as an antidote to the poison of venomous reptiles. In the treatment of chronic asthma this is an efficient remedy. The drug has a high reputation in international market and sold at high cost.

Substitutes/Adulterants of *Polygala senega*:

- ***Acalypha indica* L. (Euphorbiaceae)**. Part used : Root.
- ***Adhatoda vasica* Nees. (Acanthaceae)**. Part used : Root.
- ***Polygala chinensis* L. (Polygalaceae).** Part used : Root.
- ***Catunaregam spinosa* (Thunb.) Tirveng.. (Rubiaceae).** Part used : Root.

(V) *Saraca indica* L.*asoca* (Roxb.)deWilld.(Caesalpiniaceae).This is one of the sacred trees of Hindus and Buddhists, commonly known as 'Ashoka'. The bark of this plant has been very widely used in Ayurvedic medicine. The bark is bitter, cool astringent and used in dysentery, fever, leucorrhoea, inflammations, syphilis etc. and is help in the cure of various female diseases unfortunately this drug is subjected to a bad type of adulteration which surely stakes its merit as a therapeutic agent. Truly speaking, it is not a case of adulteration but a case of total substitution with a completely different source material. The genuine Ashoka, which they the identify as *S.asoca* is an ornamental tree mostly found only in gardens. The demand of this drug is huge due to its extensive uses in Ayurvedic

preparation like 'Ashokarishta' and extracts and tinctures, manufactured as galenicals, by the Indian Pharmaceutical concerns. Such collection is only possible if trees grow wild in abundance. But the case is not so, as stated above so obviously, it has created an easy means of substitution with quite a trash material. This fact prompted to undertake a thorough investigation of the stem bark of *S.asoca* and its Substitutes/Adulterants.

Substitutes/Adulterants of *Saraca asoca*:

- ***Bauhinia variegata* Linn. (Caesalpiniaceae).** Part used : Stem bark.
- ***Bombax ceiba* L. (Bombacaceae).** Part used : Stem bark.
- ***Polyalthia longifolia* (Sonia.) Thus. (Annonaceae).** Part used : Stem bark.
- ***Shorea robusta* Gaertn. (Dipterocarpaceae).** Part used : Stem bark.
- ***Trema orientalis* (L) Bl. (Ulmaceae).** Part used : Stem bark.

The chemistry and pharmacognosy of these plants, which are highly essential for finding out the active principles and biomarkers, are known only for a few plants or genuine drugs such as *Glycyrrhiza glabra* ,*Bergenia ligulata* and *Saraca asoca.* While very little data on phytochemistry and pharmacognosy are available for their substitutes/adulterants. Therefore the present study is undertaken to find out the active constituents and biomarkers within the main part or source of drug of all the plants where such data are not procured. In cases where the chemistry has been worked out, data are available only on their alkaloids which are considered as the active principles and in some cases, of steroids. No data on other constituents such as polyphenols and mucilages, which also may be responsible for the curative or a synergistic action are given any consideration anytime. Therefore the present project is undertaken to subject these plants to a detailed chemical, pharmacognostic, physica-chemical study and variation in the HPTLC fingerprints to detect the biomarkers which distinguishes the genuine drug from their substitutes/adulterants.

The objectives of the present Ph.D programme are the following:

1. To find out pharmacognostic biomarkers of all the substitutes/adulterants.
2. To find out active principles and chemical biomarkers of all the substitutes/adulterants.
3. To study features of purity & strength in form of ash value & extractive values of all the substitutes/adulterants.

4. To produce TLC fingerprinting of all individual substitutes/adulterants.
5. To check weather substitute/adulterant drugs are as potent as the genuine drug in terms of chemical constituents.

Chapter 2
Materials and Methods

The plant materials were collected from in and around Baroda, different parts of Gujarat and from Himachhal Pradesh, Madhya Pradesh and Mumbai. The voucher specimens of these plants are deposited in BARO, the Herbarium of Department of Botany, The Maharaja Sayajirao University of Baroda, Vadodara, Gujarat. Plant materials were washed, shade dried and later completely dried by keeping in an oven at 60 °C. The dried materials were powdered and stored in airtight plastic bags. This powder was used for the analysis of all the chemical constituents. Pharmacognostic studies (including organoleptic characterization, macroscopical and micromorphological studies) and physico - chemical analysis (The ash values and extractive values) were done by using standard methods (Anon.2004).

A. Phytochemical studies

Flavonoids:

Flavonoids are the most widely distributed group of polyphenols which include all the C_6-C_3-C_6 compounds related to a flavone skeleton .The flavone may be considered, consisting of (1) a C_6-C_3 fragment that contains the "B" ring and (2) a C_6 fragment the "A' ring, both these units being of different biosynthetic origin. The flavonoids are subdivided as anthocyanidins, flavones, flavonols, chalcones, etc. based on oxidation level of C_3 fragment of the phenyl propane unit (Geissman, 1962). These pigments sometimes completely replace the carotenoids as the yellow flower/fruit pigment. Anthocyanidins are the purple/blue pigments while chalcones and aurones are yellow in colour. Flavonols and flavones though classified as colourless flavonoids, are responsible for the white, cream or ivory colours of the flowers. All these pigments absorb strongly in ultraviolet & thus may be responsible for attracting those pollinators (e.g. bees) whose vision extends in ultraviolet region (Harborne & Smith, 1978). Flavonols, dihydroflavanols, biflavonyls, dihydrochalcones, isoflavones & proanthocynidins are the minor flavonoids since they have a restricted distribution.

'Bioflavonoids' are a group of flavonoids exhibiting pharmacological properties, especially 'Vitamin P' activity. 'Vitamin P' refers to a group of compounds which are known to be the 'permeability factors' which increase the capillary resistance and

thereby used to treat subcutaneous capillary bleeding. Rutin (3- rutinoside of quercetin), its methylated derivatives and flavanones from citrus fruits formed the principle components of Vitamin P. The interest on physiological effects of flavonoids resulted in a spurt on the research on these compounds and consequently more than 200 preparations were in use (Meyers, *et al*., 1972). It is experimentally established that flavonoids with free hydroxyl groups at the 3', 4'- positions exert beneficial physiological effects on the capillaries through (1) chelating metals and thus sparing ascorbate from oxidation, (2) prolonging epinephrine action by the inhibition of O-methyl transferase and (3) stimulating the pituitary-adrenal axis (De Eds, 1968). Srinivasan *et al*., (1971) presented evidence that flavonoids play another important role in circulatory system by acting on the aggregation of erythrocytes.

Most of the flavonoids occur as water soluble glycosides in plants. They are extracted with 70% ethanol or methanol and remain in the aqueous layer, following partition of this extract with solvent ether. Due to the phenolic nature of flavonoids, they change in colour when treated with bases (esp. ammonia) and thus are easily detected in chromatograms or in solutions. Flavonoids contain conjugated aromatic system and thus show intense absorption bands in UV and in the visible regions on the spectrum. A single flavonoid aglycone may occur, in a plant, in several glycosidic combinations and for this reason it is considered better to examine the aglycones in hydrolysed plant extracts (Harborne, 1984).

Normally the flavonoids are linked to sugar by *O*-glycosidic bonds, which are easily hydrolysed by mineral acids. But there is another type of bonding in which sugars are linked to aglycones by C-C bonds. The latter group of compounds, known as C-glycosides (glycoflavones), are generally observed among flavones. They are resistant to normal methods of hydrolysis and will remain in the aqueous layer when the hydrolysed extract is extracted with ether to remove aglycones.

The procedures followed in the present work for the extraction, isolation and identification of flavonoids are described below.

Fifty grams of leaf power was extracted in a Soxhlet's apparatus with methanol for 48hrs till the plant material became colourless.The methanolic extract was concentrated to dryness in a water bath. 25-30 ml of water was added to the dry residue and the water soluble phenolic glycosides were filtered out. The filtrate was hydrolysed in a water-bath for one hour using 7% HCl. This hydrolysate was extracted with diethyl ether/solvent ether, whereby the aglycones got separated into

ether fraction (fraction A). The remaining aqueous fraction was further hydrolysed for another 10 hours to ensure the complete hydrolysis of all the O-glycosides. Aglycones were once again extracted into diethyl ether (fraction B) and the residual aqueous fraction was neutralized and evaporated for the analysis of glycoflavones.

Ether fractions A and B were combined and analysed for aglycones using standard procedures (Harborne, 1967, 1984; Mabry *et al*; 1970; Markham, 1982). The combined concentrated extract was banded on Whatman No. 1 paper & chromatographed along with quercetin as the reference sample. The sample system employed were Forestal (Con.HCl: acetic acid: water; 30:30:10) or 30% glacial acetic acid. The developed chromatograms were dried in air and the visibly color compounds were marked out. These chromatograms were observed in the ultra-violet light (360 nm) and the bands were noted. Duplicate chromatograms were then sprayed with 10% Na_2CO_3 and 1% $FeCl_3$ and the color changes were reported. Rq (Rf relative to quercetin) values were calculated for all the compounds. The bands of the compounds were cut out from unsprayed chromatograms and were eluted with spectroscopic grade methanol. The UV absorption spectra of these compounds were recorded in methanol using Perkin-Elmer Lambda 25 UV/Vis spectrophotometer. The bathochromic & hypsochromic shifts induced by the addition of various regents were studied. The reagents used and there preparation are given below (Mabry *et al*; 1970).

Sodium methoxide (NaOMe): Freshly cut sodium metal (2.5) gm was added cautiously in small portion to spectroscopic methanol (100ml). The solution was stored in a tightly closed glass bottle.

Aluminium chloride ($AlCl_3$): Five grams of fresh anhydrous AR grade $AlCl_3$ (which appeared yellow-green and reacted violently when mixed with water) were added cautiously to spectroscopic methanol (100ml), formed initially, dissolved after about 24 hrs.

Hydrochloric acid (HCl): Concentrated AR grade HCl (50ml) was mixed with distilled water (100ml) and the solution was stored in glass stoppered bottle.

Sodium acetate (NaOAc): Anhydrous powdered AR grade NaOAc was used.

Boric acid (H_3BO_3): Anhydrous powdered AR grade $H_3BO_3$was used.

The concentrations of the sample solution prepared by eluting chromatogram strips were adjusted so that the optical density (OD) fell in the region of 0.6 to 0.8. The methanol spectrum

was taken using 5 ml of this stock solution. A reference solution was prepared by extracting a piece of blank chromatographic paper from the same chromatogram with spectroscopic methanol. The NaOMe spectrum was measured immediately after the addition of three drops of NaOMe stock solution to the flavonoidal solution used for methanol spectrum. The solution was then discarded. The $AlCl_3$ spectrum was then measured immediately after the addition of six ml of $AlCl_3$ stock solution to 5 ml of fresh stock solution of the flavonoids. $AlCl_3$ /HCl spectrum was recorded next, after the addition of 3 drops of the HCl solution to the solution containing $AlCl_3$. The solution was then discarded. For NaOAc spectrum, excess coarsely powdered anhydrous AR grade NaOAc was added by shaking the cuvette containing 5 ml of fresh solution of the flavonoids, till about a 2mm layer of NaOAc remained at the bottom of the cuvette. The spectrum was then recorded 2 minutes of the addition of NaOAc. NaOAc/ H_3Bo_3 spectrum was taken after sufficient H_3Bo_3 was added to give a saturated solution. The solution was discarded after recording the spectrum. The structure of flavonoid was established by its absorption maxima (λ_{max}), shape of the curves, shifts (both bathochromic & hypsochromic) with different reagents $AlCl_3$, $AlCl_3$ /HCl, NaOAc/H_3Bo_3, NaOMe (Mabry et al; 1970), color reactions and Rf values. The identification was confirmed by co - chromatography with authentic samples.

The procedures followed for isolating glycoflavones are described below:

The aqueous fraction remaining after the separation of agylcones was neutralized by the addition of anhydrous Na_2CO_3 / $BaCO_3$ and concentrated to dryness. When $BaCO_3$ was used, barium chloride got precipitated and was filtered out. This filtrate was concentrated to dryness. To this dried residue, ethanol was added to dissolve the glycoflavones. The alcoholic filtrate was concentrated, and was banded on Whatman No.1 paper and the chromatogram was developed in water as solvent system. Glycoflavones were visualized by their colour in UV & with 10% aqueous Na_2CO_3 spray. Further analysis and identification were done by measuring the λ max and spectral shifts and co-chromatography with authentic samples.

Phenolic acids:

Phenolic acids are simple phenols, having a functional acidic group and varying number of hydroxyl groups at different position. Acid hydrolysis of plant tissue releases a number of ether-soluble phenolic acids, some of which are universal

in distribution. These acids occur either associated with lignin or are bound to the glycosides. They are also seen as depsides or as esters in hydrolysable tannins. Ellagic acid and gallic acid are located in many plant groups of the Polypetalae. The phenolic acids are extracted in ether along with the flavonoid agylcones from the hydrolysed extract (fraction A and B) of plant materials. They are analysed as follows:

Analysis of phenolic acids in the combined ether fraction (A and B) was carried out by two-dimensional ascending paper chromatography. Benzene: acetic acid: water (6:7:3, upper organic layer) in the first direction and sodium formate: formic acid: water (10:1:200) in the second direction were used as irrigating solvents. The sprays used to locate the compounds on the chromatograms were diazotized *p*-nitraaniline or diazotised sulphanilic acid and a 10% Na_2CO_3 overspray (Ibrahim and Towers, 1960).
Diazotization: 0.7gms of *p*-nitraniline/sulphanilic acid was dissolved in 9 ml of HCl and the volume made up to 100 ml. Five ml of 1% $NaNO_2$ was taken in a volumetric flask and kept in ice till the temperature was below 4°C. The diazotized sprays were prepared by adding 4 ml of *p*-nitraniline/sulphanilic acid stock solution to the cooled $NaNO_2$ solution. The volume was made up to 100 ml with ice-cold water.

The various phenolic acids presents in the extract were identified based on the specific colour reactions they produce with the spray reagents and the relative Rf values in the different solvent system.

Quinones:

They are aromatic diketones, which form the largest class of natural coloring matters. They are generally known from higher plants and fungi. In higher plants they play a subsidiary or a secondary role. They are generally present in the bark or underground parts. In leaves their color is masked by other pigments. They are classified into benzo-, naptha- and anthraquinones depending on the mono-, bi- or tricyclic ring system they contain. In plants their function is not properly understood. It is assumed that they play some role in oxidation-reduction processes.

For extraction of quinones, approximately 5-10 gm of dried, powdered 5-10 gm of dried, powdered leaf material was exhaustively extracted with hot benzene for 3 x 12 hrs and the extract was dissolved in solvent ether and segregated into acidic and neutral fractions by repeatedly shaking with 2N Na_2CO_3 solution. The Na_2CO_3 soluble fraction was acidified with ice-cold 2N HCl dropwise till the precipitate formed settled down. The acidified solution, in turn, was extracted with diethyl ether

and separated again into two layers. The lower layer was discarded, while the upper acidic fraction was chromatographed over TLC (silica gel G) plates using petroleum ether-benzene (9:1) as the solvent system (Joshi *et al.*, 1973).

The neutral fraction was also chromatographed over silica gel TLC plates using the same solvent system. The various quinones (Anthra-, Benzo-, Napthaquinones) were visualized by their colours in visible/UV light, colour reactions after spraying with 2% magnesium acetate or 10% aqueous NaOH (the quinones give purple/pink/orange-yellow colours) and the absorption spectra.

Proanthocyanidins:

The proanthocyanidins are condensed tannins which yield anthocyanidins on hydrolysis. For testing the presence of proanthocyanidins, about 5 gm of finely chopped (fresh) leaf material or 2 gm dry powdered material was taken in 20 ml test-tube and covered with approximately 5 ml of 2N HCl. Extraction was carried out by placing the test-tube in a boiling water bath for half an hour. The extract was decanted after cooling and shaken with amyl alcohol. Presence of a red or near carmine color in the upper alcohol layer denoted a positive reaction for proanthocyanidins. An olive yellow color represented a negative reaction (Gibbs, 1974). The colored hydrolysate is extracted with amyl alcohol and this extract was chromatographed in Whatman No.1 paper using Forestal or 30% HOAc. The anthocyanidins which separate as colored bands were eluted with acidic methanol and the absorption spectra were measured in the range between 500-600nm. The different anthocyanidins were identified by their visible colors, Rf values and $\lambda_{max.}$

Alkaloids:

Alkaloids comprise the largest single class of secondary metabolites. They are basic plant products having nitrogen-containing heterocyclic ring system & a high pharmacological activity. Alkaloids, as a rule, are insoluble in water but soluble in organic solvents. But their salts are soluble in water and insoluble in organic solvents Alkaloids are normally extracted from plants into weakly acids (1M HCl or 10% acetic acid) or acidic alcoholic solvents and are then precipitated with alcoholic ammonia. They are also extracted into any organic solvents after treating plant material with a base. The base frees the alkaloids and makes them soluble in organic solvents. From the organic solvents, the alkaloids are extracted into acidic solution and tested with specific regents.

Five grams of powdered plant material was cold extracted with 50 ml of 5 % ammoniacal ethanol for 48 hours. The extract was concentrated (by distillation and the residue was treated with 10 ml of 0.1N H_2SO_4. The acid soluble fraction was tested with Mayer's, Wagner's and Dragendorff's regents (Paech and Tracey, 1955). The white precipitate denoted the presence of alkaloids. The acid soluble fraction was spotted on TLC (Toluene: EtoAc: diethylamine; 7:2:1) and the Rf values are measured. The preparation of the reagents was as follows:

Mayer's reagent: (Potassium mercuric iodide) 1.36gm of $Hgcl_2$ were dissolved in 60 ml of distilled water and 5gm of KI in 10ml of solvent. A few drops only of this reagent were added, as precipitates of some alkaloids were soluble in excess of the reagent**.**

Wagner's reagent: (Potassium iodide) 1.27 gm of I_2 and 2gm of KI were dissolved in 5 ml of water and the solution diluted to 100 ml. It gave brown flocculent precipitate with most of the alkaloids.

Dragendorff's reagent: (Potassium bismuth iodide) 8gm of $Bi(NO_3)_3.5H_2O$ were dissolved in 20 ml of HNO_3(sp. gr. 1.18)and 27.2 gm of KI in 50 ml of water. The two solutions were mixed and allowed to stand when KNO_3 crystallized out. The supernatant was decanted off and made up to 100 ml with distilled water.

The extract which showed presence of alkaloids were concentrated and this fraction was spotted in Whatman No.1 chromatographic papers along with standard alkaloids such as ephedrine, berberine, quinine etc. and developed in BAW. The developed chromatograms were seen in UV light and the fluorescent regions were marked. The chromatograms were then sprayed with Dragendorff's reagent

Gums and Mucilages

Gums and mucilages include all the hydrocolloids obtained from plants and are polysaccharides consisting of more than one type of monosaccharide residues. The gums are considered as pathologic products, produced in response to injury by a process known as " gummosis", whereby the cell walls and their ingredients are dissolved to form a colloid which serves as a protective layer over the wounded tissue and later occurs as exudates from the various plant parts especially the trunk. Mucilages, on the other hand, are classified as natural plant products produced by the plant for the imbibition and retention of water. But from a chemical point of view gums and mucilages are almost identical and it is nearly impossible to draw a line

demarcating one from the other.

The solutions of gums and mucilages are laevorotatory. On hydrolysis they yield sugars like arabinose, galactose, glucose, mannose and xylose alongwith various uronic acids and methyl sugars. The sugar acids when present in appreciable amounts, tend to lower the pH of the solution enabling the gums to occur frequently as salts of sodium, potassium, calcium or magnesium. In some cases, the sugar components are methylated (gum Tragacanth) or acetylated (Karraya gum). The trace amounts of nitrogen (0.08-5.6%), at times encountered in certain samples of gum, are considered to be due to the presence of proteins or sugar amines like glucosamine.

Gums containing linear polysaccharides are found to be less soluble in water, producing very viscid solutions. They tend to precipitate in course of time because of the inter- molecular hydrogen bonding facilitated by the parallel alignment of molecules. The solutions of branched polysaccharides are more soluble in water and form colloidal gels-sols-possessing low surface tensions and therefore act as important protective colloids and stabilising agents.

B. Pharmacognostic studies

1. Organoleptic characterization

The organoleptic evaluation comprise macroscopic and sensory characteristics of drugs. The purview of study comprises morphological origin, condition, shape, size, colour, texture, taste, odour, hardness and fracture (Wallis, 1957).

2. Micromorphology and Anatomy

Micromorphological and anatomical studies were carried out on fresh materials. Fresh leaves were washed and small fragments of leaves were taken from the middle region of the mature leaves. Washed leaf fragments were first boiled in 90% alcohol for about 3-5 minutes to remove chlorophyll, then washed 2-3 times in water, then again boiled with 10% KOH solution (Wallis, 1957) for 2-3 minutes and washed 4-5 times in water and kept in clean water to remove all traces of the clearing agent. Both the epidermal layers were stripped off gently from the mesophyll tissue with their help of pointed needle and forceps. The epidermal peels were washed in water, stained with Toluidine blue (0.5%) prepared in aqueous borax (Trump, 1961) and mounted in 50% glycerine; the margins of the cover slips were sealed with DPX (Johansen, 1940). Transverse sections of leaf, as well as T.S., T.L.S and R.L.S of stem

and roots were taken by free hand and were stained in Toluidine blue (0.5%) and mounted in 50% glycerine. The slides were examined under the microscope and Camera Lucida sketches were drawn at 400x magnification and the size was measured using an ocular micrometer. The quantitative data were based on the average of 20 readings.

Leaf constants such as stomatal index/mm^2 and trichome index/mm^2 were calculated. Stomata index (SI) was calculated as defined by Salisbury (1927, 1932) *viz.*

$$SI = \frac{S}{S + E} \times 100$$

3. Powder study

powdered drugs explain cellular element of respective morphological parts and their inclusions. The powder of the whole drugs consists of the elements of all the morphological parts included in the drugs. The finely powdered drug was scanned under 400x magnification for recording the cell elements.

C. Physico-chemical analysis

There are certain physico-chemical parameters viz. Total Ash, Acid Insoluble Ash, Alcohol Soluble Extractive and Water Soluble Extractives. The procedures followed for the determination of the various parameters during proximate analysis are as follows (Anon.2004).

1. Total ash content

A silica crucible was heated to red heat for 30 minutes and was allowed to cool in a dessicator. The crucible was weighed. Accurately 2 grams of the air-dried plant powder was weighed into the crucible and was incinerated at a temperature not exceeding 450°C until free from carbon. The crucible was cooled and weighed. The percentage of total ash formed was calculated with respect to the air-dried plant powder.

2. Acid insoluble ash content

The total ash obtained from the above procedure was boiled with 25 cc of 2M HCl solution for 5 minutes. The insoluble matter was collected on an ashless filter paper, washed with hot water, ignited in a crucible and cooled in a dessicator. The weight

was noted. The percentage of the acid insoluble ash was calculated with reference to the air dried plant powder.

3. **Alcohol soluble extractive**

Five grams of air-dried plant material was macerated with 100cc of alcohol in a closed flask for 24 hours, shaking frequently during the first 6 hours and allowed to stand for the remaining 18 hours. Thereafter, filtration was done rapidly taking precautions against loss of ethanol. Twenty five cc of the filtrate was evaporated to dryness in a tared flat bottomed shallow dish, dried at 105°C and weighed. The percentage of the ethanol soluble extractive was calculated with respect to the air-dried plant powder.

4. **Water soluble extractive**

Chloroform water was prepared by dissolving 2.5cc of chloroform in 900cc of distilled water and diluting upto 1000cc with water. Using the prepared chloroform water, the procedure followed was similar to that of the ethanol soluble extractive. The percentage of the water soluble extractive was calculated with respect to the air-dried plant powder.

Chromatographic Characterization

Chromatography is the science which studies the separation of molecules based on differences in their structure and/or composition. In general, chromatography involves moving a test preparation of the materials to be separated, over a stationary support. The molecules in the test preparation will have different interactions with the stationary support leading to separation of similar molecules. Test molecules which display tighter/ stronger interactions with the support will tend to move more slowly through the support than those molecules with weaker interactions. In this way, different types of molecules can be separated from each other as they move over the support material. Chromatographic separations can be carried out using a variety of supports, including immobilized silica on glass plates (thin layer chromatography), very sensitive High Performance Thin Layer Chromatography (HPTLC), volatile gases (gas chromatography), paper (paper chromatography), and liquids which may incorporate hydrophilic, insoluble molecules (liquid chromatography).

D. HPTLC finger-printing Analysis

Regulatory agencies recommend fingerprint chromatography as the basis for proper identification of herbal drugs, herbal drug preparations and herbal medicinal products. In general, the fingerprint of one sample (unknown) is compared with that of another sample (i.e. reference material). The reference material can either be of the same kind as the unknown (herbal drug or preparation thereof) or be a solution of any number of chemically defined substances. Fingerprints are compared with respect to number, sequence, position and colour of the separated zones. The fingerprint can be optimized for certain target compounds. Even if some components migrate with the solvent front and others remain at the application position, the fingerprint always represents the sample in its entirety.

A balance must be found between the number and type of compounds extracted, and those of interest, because unwanted substances can disturb the analysis. Proteins, lignans and sugars can constitute an undesirable matrix. In cases where the active principles are known, they can serve as markers. If no active principle has been determined, any of the secondary metabolites like essential oils, flavonoids, alkaloids, or others which are in appreciable quantities, may be selected. Even amino acids, plant acids or sterols can provide plant-specific profiles. There could be cases where little or nothing is known about the chemical constituents of a given herbal drug. In such cases, generating multiple fingerprints is very valuable. Multiple fingerprints can be obtained either from the same plate by multiple detections or from the same sample looking at different fingerprints representing various substance classes.

The traditional way of describing an HPTLC fingerprint chromatogram is comparison of the sequence, colour and intensity of the separated zones of the sample, with that of the reference. The use of chemically defined compounds as reference material may be preferred, because such compounds are readily available in suitable purity. Only against such substances can zones of the HPTLC fingerprint be identified, unless the unknown is isolated and externally analyzed. Such reference materials also provide the option of estimating the quantitative composition of the sample.

Here in this work HPTLC analysis done qualitatively aim to develop HPTLC fingerprint for individual drugs for comparison and to distinguish genuine drugs from their substitutes/adulterants.

Preparation of extracts for HPTLC analysis

One gm of coarse powder material were extracted by refluxing in 5ml of methanol at 60°C in a water bath for 30 min. Extracts were filtered, concentrated and re-suspended in 1ml of methanol and used directly for HPTLC analysis.

Table 1: Optimized chromatographic conditions for HPTLC analysis.

HPTLC Sample applicator	Linomat 5 (CAMAG)
Make of syringe	Hamilton
Capacity of syringe	100µL
Development chamber	CAMAG twin trough chamber (10x10cm)
Stationary phase	Precoated $60F_{254}$ silica plates (Merck)
Size of plate used	10x10 cm
Sample applied	10 µl
Distance between tracks	12 mm
Band length	8mm
Solvent front	90 mm
Mobile phase	Toluene :Formic acid : Ethyl formate (5 : 1 : 4) (v/v/v)
Mode of visualization	Short wave UV light (UV 254nm) Long wave UV light (UV 366nm)
HPTLC scanner	TLC Scanner 3 (CAMAG)
Radiation source	Deuterium lamp
Software	WinCats

Chapter 3

a. *Fumaria parviflora* Lam.(Fumariaceae)

Synonyms: *Fumaria indica* (Hausskn.) Pugsley.

Sanskrit: Parpata, Arako, Charaka, Pittari.

Vernacular names:

Arabic : Baglatulmulk, Bukslatulmulik, Shahatraja.

Bengali: Bansulpha ,Shotara pipapapra.

English : Common fumitory, Fine-leaved Fumitory.

Gujrati : Pittapapdo.

Hindi : Pitpapra, Pitpapada, khetpapra.

Kannad : Parpataka.

Marathi : Pittapapra.

Sindhi : Shahatra, Shatra.

Tamil : Turu, thusha.

Telgu : Chata-rashi, Chatarasi.

Urdu : Shahatra.

Distribution and habitat

The plant is a small, scandent, branched annual herb distributed throughout India, growing wild in plains and lower hills particularly on the banks of the Ganges and in the Himalayas up to an altitude of 2700 m. It is also found in Europe, Africa and many other Asian countries.

Morphological features

The plant is a pale green much branched up to 2 ft. height, an annual herb, suberect or diffuse. Leaves multifid more of less glaucous; leaflets 2-4 pinnatisect; segment long, linear or linear-oblong, flat, acute. Recemes with 10-12 flowers rather dense in flower, bract, lanceolate-subulate, slightly acuminate, pedicels 2-2.5, rarely 4.5 mm long, erect thickened at the apex. Sepals about 1.5 mm long, 0.5-1 mm broad, lanceolate or ovate, acuminate more or less inciso- dentate, rose colored often persistent in the young fruit. Corrola 5-6 mm long rose colored. Fruit about 2.5 mm, broad, subrotund, quadrate, subtrucate and sometimes obscurely retuse. Stem light green, smooth hollow about 3-4 mm thick, root brown color, branched about 2-3 mm thick, cylindrical.

Medicinal uses

The whole plant is widely used in traditional and folkloric systems of medicine. The plant is regarded as a laxative, diuretic, diaphoretic and is beneficial in dyspepsia, liver complaints and scrofulus skin affections(Kirtikar and Basu, 1985)and used in fever, influenza(Anon. 1956), syphilis, scrofula, leprosy, constipation, ague and jaundice. The decoction of stem and leaves is gives as a tonic, anthelmintic , aperients (Rastogi and Mehrotra, 1970-79) and claimed to possess various curative properties for ailments of the blood, skin, gastrointestinal system and central nervous system(Usmanghani,1997).

Previous Phytochemical reports

The plant is found to contain potopine, tetrahydro coptisine, tautomeric form of fumariline, a homogenous gum, a racemic mixture of bicuculine and its optical antipode, bicuculine, fumarilicine and narceimine (Pandey *et al.*,1971). Later on protopine, quanternary salt of protopine, nona cosanol and sitosterol were isolated from the stem and leaves of *F. indica* (Satish and Bhakuni.1972). fumariline, 8-methoxy dihydro sanguinarine and oxysanguinarine (Pandey, Gupta and Ray.1979). A secopthalide isoquinoline alkaloid narceimine isolated from seeds (Pandey *et al.*,1988). Isoquinoline base papracine along with six known base oxyhydrastinine, noroxyhydrastinine, fumaramine, stylopine, bisnorargemonine and fumariti(Rahman, Bhatti and Choudhary.1992). Two new spirobeanzyl isoquinoline (tyramine base) alkaloids, papracinine and paprazine together with six other known alkaloids as fumaritine N-oxide, parfumine, lastourvilline, feruloyl tyramine, fumariflorine and N-methyl corydaldine identified from aerial parts of *F. indica* (Rahman *et al.*,1992) A new seco-phalidi isoquinoline alkaloid narlumicine from stem of *F. indica* together with protopine nitrate, protopine, DL - tetrahydrocoptisine and narlumidine have been reported (Pandey and Tripathi 1992) Similarly three new seco-pthalide isoquinoline alkaloids peprafumine, peprarine and papraline along with three other known alkaloids cryptopine, raddeanine and oxocoptisine have been identified from the aerial part of *F. indica* (Rahman *et al.*,1995). Recently, a new alkaloid, fuyuziphine together with (+/-)-alpha-hydrastine has been isolated from the whole plant of *F. indica* (Pandey *et al.,* 2008).

Previous pharmacognostic reports

Very little data available on pharmacognosy of this plant. Only T.S of various parts of the plant has been studied (Anon.2004). In the present work roots, stem and leaves of this plant has been subjected to phytochemical and pharmacognostic studies.

Materials and methods

The plant material has been collected from Tarikhet, Uttaranchal. Phytochemical analysis of the plant for their secondary metabolites such as alkaloids, flavonoids, phenolics, steroids, quinones, mucilages and other compounds were done by standard methods of chromatography and spectrophotometry described in Chapter 2. Pharmacognostic, HPTLC finger printing and physico-chemical studies were done by using standard methods described in Chapter 2.

Results

Phytochemistry

The plant is found to contain flavonol quercetin and 3'-0Me quercetin. The phenolic acids located were vanillic and ferulic (*cis*- and *trans*- isomers) acids while syringic acid was found absent. Mucilage amounted to 6.3 % consisting of ribose and xylose. The plant also showed the presence of unidentified alkaloids, steroids and tannins.

Pharmacognosy

Root : T.S (Fig.1)

The T.S. was circular in outline. Cork were poorly developed and cells were 2 to3 layered. The cells were arranged one above the other. The cortex was 6-11 layered of compactly arranged thin walled cells. The cells of outer 2-4 layers were small and square while inner ones were large polygonal and bearing light brown contents.. The vascular bundle characteristically was fan shaped. The secondary phloem was 6-8 layered followed by wide zone of central wood. The rays were poorly developed and were uni- to biseriate. Some of them also were filled with light brown contents. The xylem vessels were many in number, mostly occurred singly but few were in groups of 2 were almost uniform in size. The xylem vessels were surrounded by fiber tracheids having large lumen. The primary xylem was not distinct.

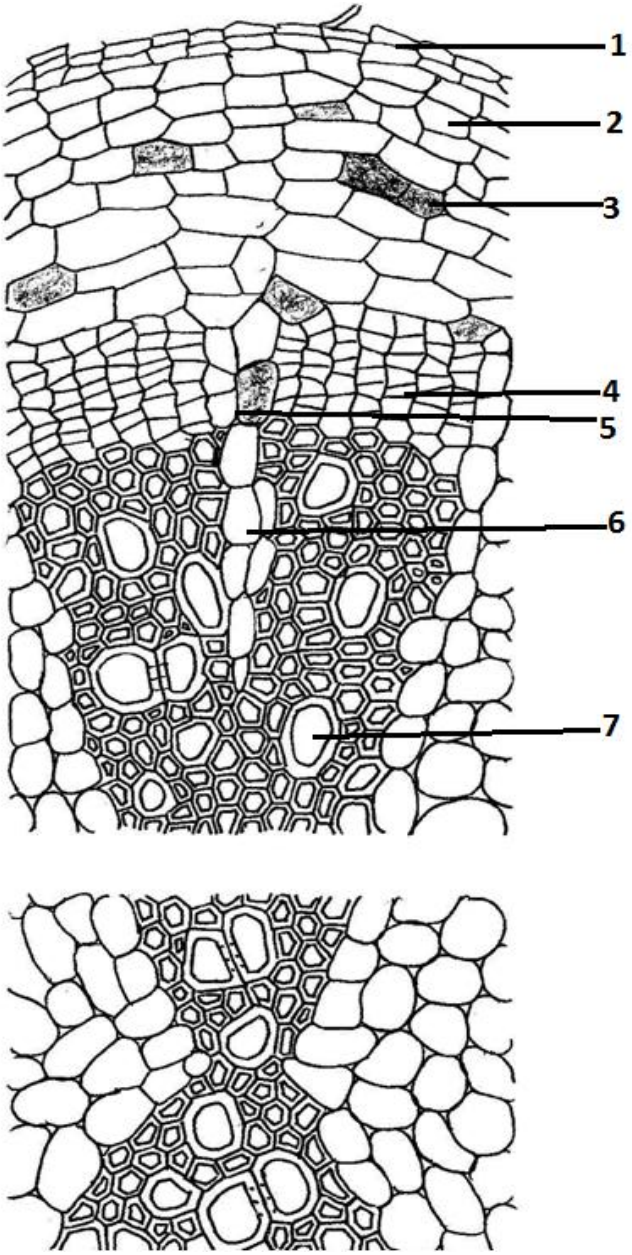

Fig.1.*Fumaria parviflora* root, T.S: 1. Cork, 2. Cortex, 3. Cortical parenchyma bearing light brown contents, 4. Phloem, 5. Phloem ray, 6. Xylem ray, 7. Vessel.

Root : T.L.S (Fig. 2)

The cells of the cortex were rectangular thin walled, some of bearing light brown contents. The fibers were curved around the spindle shaped medullary rays and each ray cell was thin walled and polygonal. Fiber tracheids were pitted, and contained simple pits in them. The vessels were broad, reticulate and bordered pitted. Scalariform thickened vessels were also observed.

Root : R.L.S (Fig.3)

The phloem parenchyma cells were erect, rectangular thin walled to which 3 to 4 companion cells were attached . Xylem parenchyma were homogenous upright, square and found with simple pits. Wood fibers were straight.

Leaf micromorphology

The stomata were of anomocytic type. The stomatal index was 15-18. Trichomes were found absent.

Leaf : T.S (Fig.4)

In the **lamina portion** there were stomata on the both the sides. The cells of epidermis were barrel shaped and covered by a thin cuticle. The mesophyll was not differentiated into palisade and spongy parenchyma and consisted of compactly arranged (having very little intercellular spaces)thin walled polygonal parenchyma filled with chlorophyll. vascular bundled were scattered throughout the mesophyll.

Stem : T.S (Fig.5)

The T.S. was quadrangular to pentagonal in outline and showed a thick cuticle covered the barrel shaped cells of the single layered epidermis. Cortex was of 3-5 layers of thin walled parenchyma cells, most of the cells contained chlorophyll and became chlorenchymatous and collenchymatous at the portion below ridges. The cells of collenchyma were comparatively small in size. Endodermis was indistinct. The vascular bundles were in a ring present either single or in group of 2-3 and found present below the ridges. Phloem zone was covered with sheaths of sclerenchyma. The cells of sclerenchyma were of two types, small, rounded without striations and big, slightly oblong with striations. The phloem was 4-6 layered and consisted of usual elements. Xylem consists of usual elements. Vessels were mostly having simple and reticulate thickening were as spiral or annular thickening found occasional. wood fibers were 3-4 in a group. The pith was parenchymatous outer and hollow in the centre leaving a cavity. The cells were thin walled and were loosely arranged.

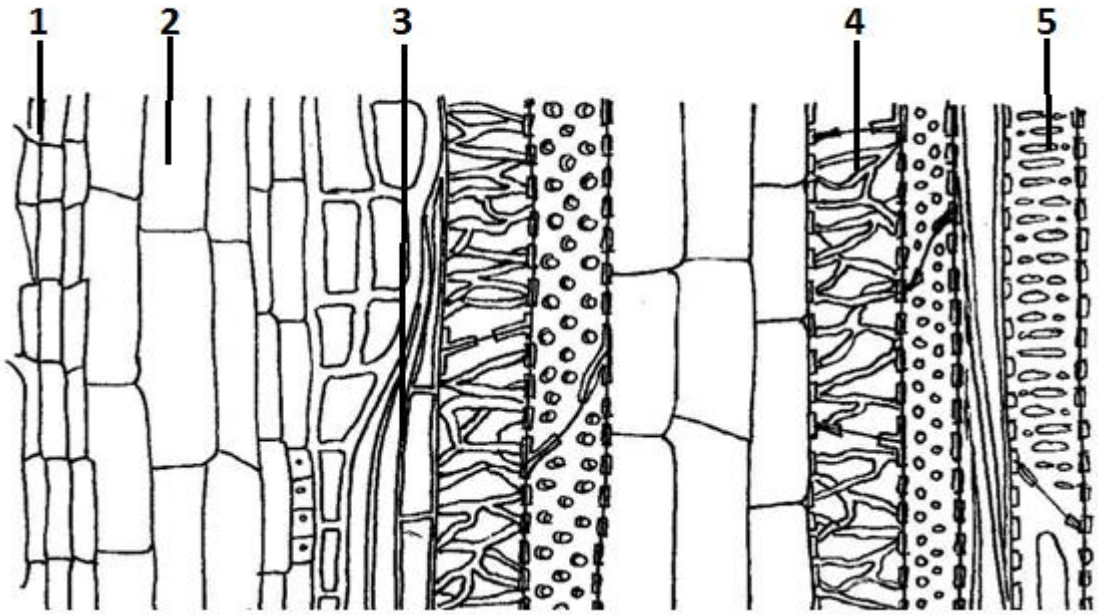

Fig.2.*Fumaria parviflora* root, T.L.S: 1.Cork, 2.Cortex, 3.Wood fiber, 4. Reticulate vessel, 5. Scalariform vessels.

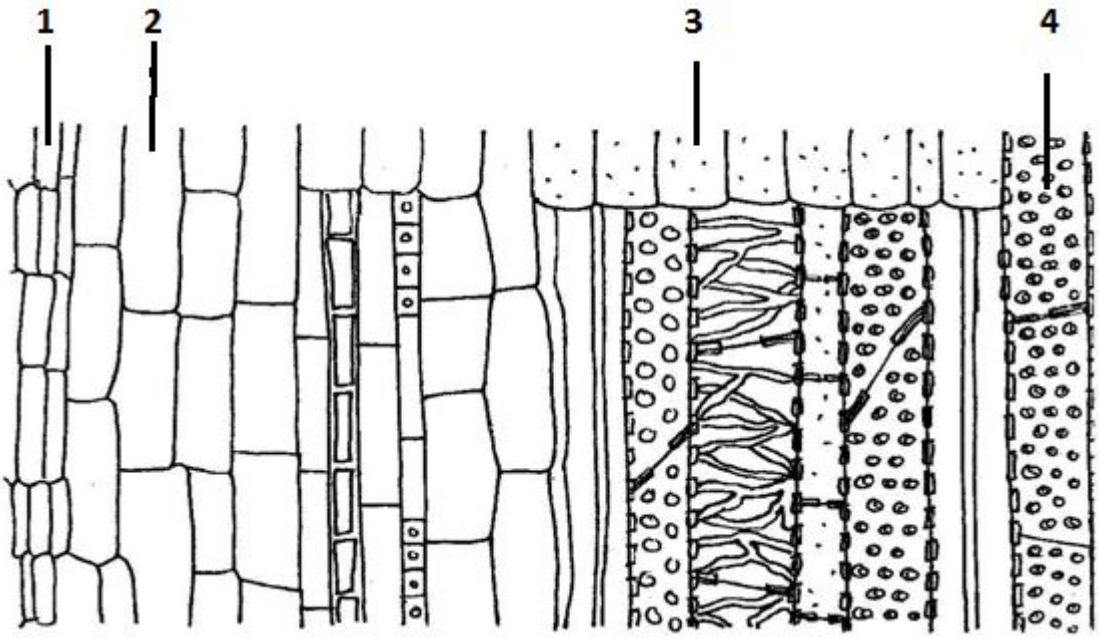

Fig.3.*Fumaria parviflora* root, R.L.S:1. Cork cells, 2.Cortex, 3. Xylem rays, 4.Boarded pitted vessel.

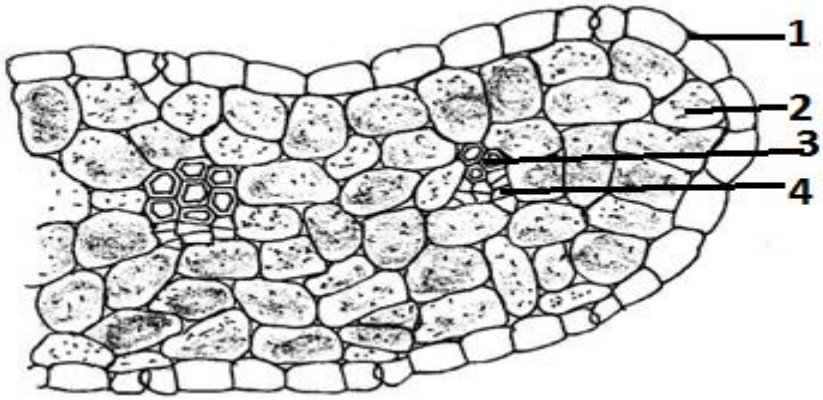

Fig.4.*Fumaria parviflora* leaf,T.S:1.Epidermis,2. Mesophyll, 3. Xylem, 4.Phloem.

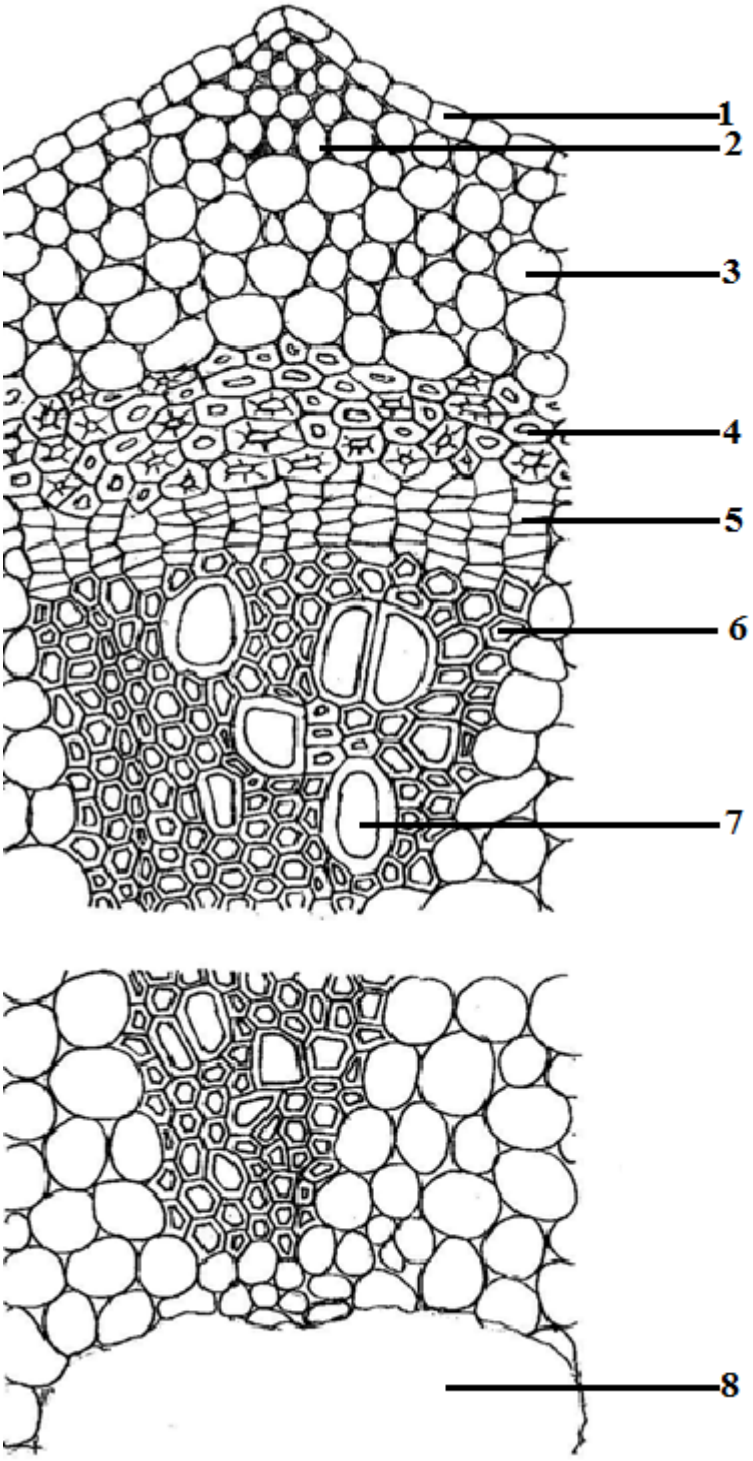

Fig.5.*Fumaria parviflora* stem, T.S : 1.Epidermis, 2.Collenchyma, 3.Cortex, 4.Sclerenchyma, 5. Phloem, 6. Xylem, 7. Vessel, 8. Centre hollow pith.

Stem : T.L.S (Fig.6)

The cortical parenchyma cells were large rectangular contained chlorophyll followed by a group of sclerenchyma. Vessels showed simple and reticulate thickening. Xylem parenchyma were square and found with simple pits. The cells of pith were thin walled and large polygonal.

Stem : R.L.S (Fig.7)

Epidermal cells were barrel shaped followed by rectangular cells of collenchyma. Wood fiber were with simple pits. Vessels showed simple, reticulate and annular thickening. Pith showed large cavity.

Powder study (Fig.8)

The components present in the powder were cork, fragments of collenchyma, parenchyma with light brown deposits, sclerenchyma, septet fibers, epidermal fragments with stomata and reticulate vessels.

Distinguishing features

Pharmacognostic markers :

Root

1. Cortical parenchyma cells bearing light brown contents.
2. Fan shaped vascular bundle.
3. Pitted fiber tracheids.

Leaf

1. Deposition of cystoliths and globules in the epidermis.
2. Anomocytic type of stomata.
3. Mesophyll was not differentiated into palisade and spongy parenchyma
4. Absence of trichomes.

Stem

1. Pith showed large central cavity.
2. Vascular bundle was capped with sclerenchymatous sheath.
3. Vessels showed reticulate thickening.

Phytochemical markers

1. Quercetin.
2. 3'-OMe quercetin.
3. Vanillic acid.
4. Ferulic (*cis*- and *trans*- isomers) acid.

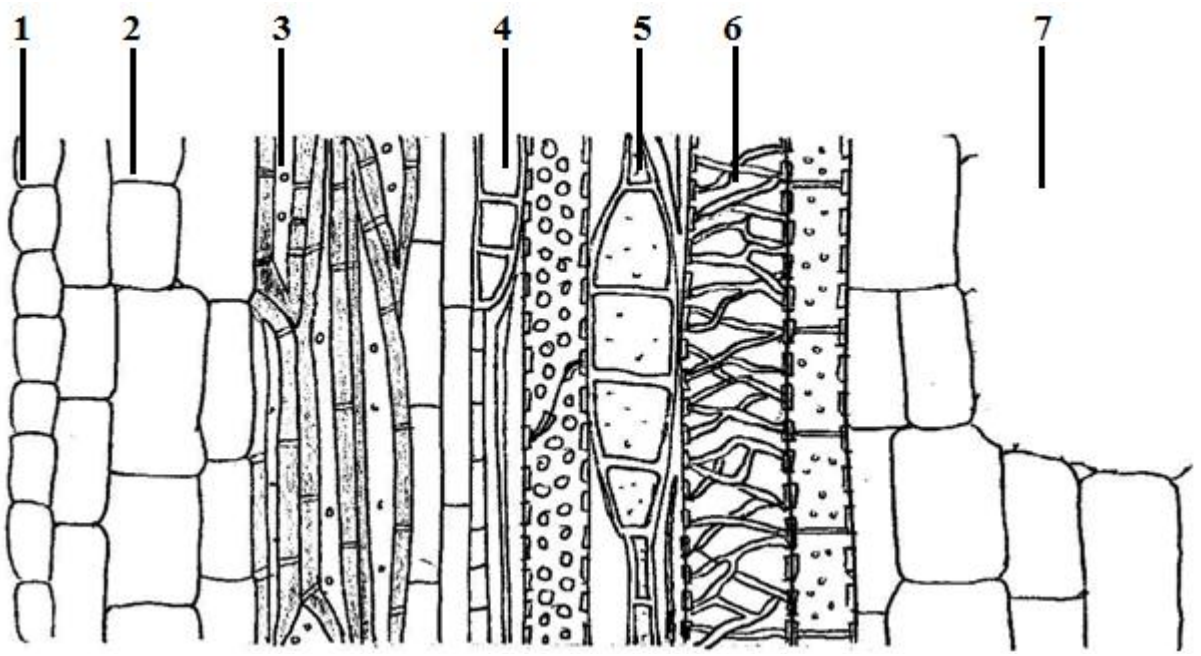

Fig.6. ***Fumaria parviflora*** **stem, T.L.S**:1. Epidermis, 2. Cortex, 3. Sclerenchyma, 4. Phloem ray, 5. Xylem rey, 6. Reticulate vessel, 7. Centre hollow pith.

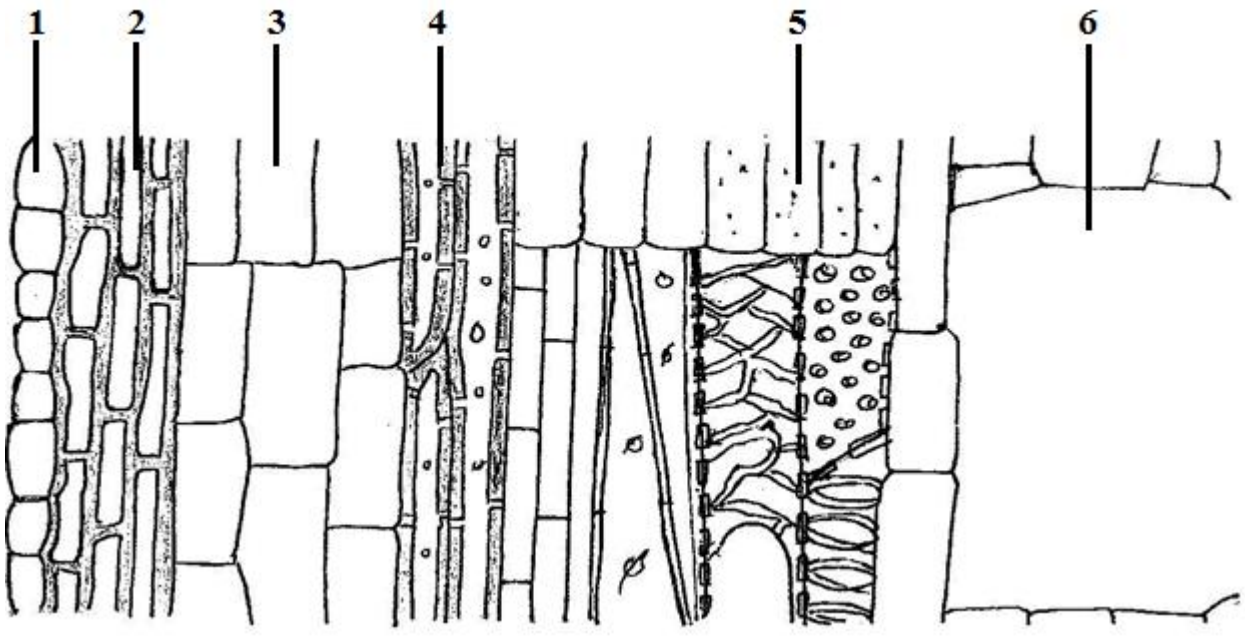

Fig.7.*Fumaria parviflora* **stem, R.L.S**: 1. Epidermis, 2. Collenchyma, 3. Cortex, 4. Sclerenchyma, 5. Xylem rey, 6. Centre hollow pith.

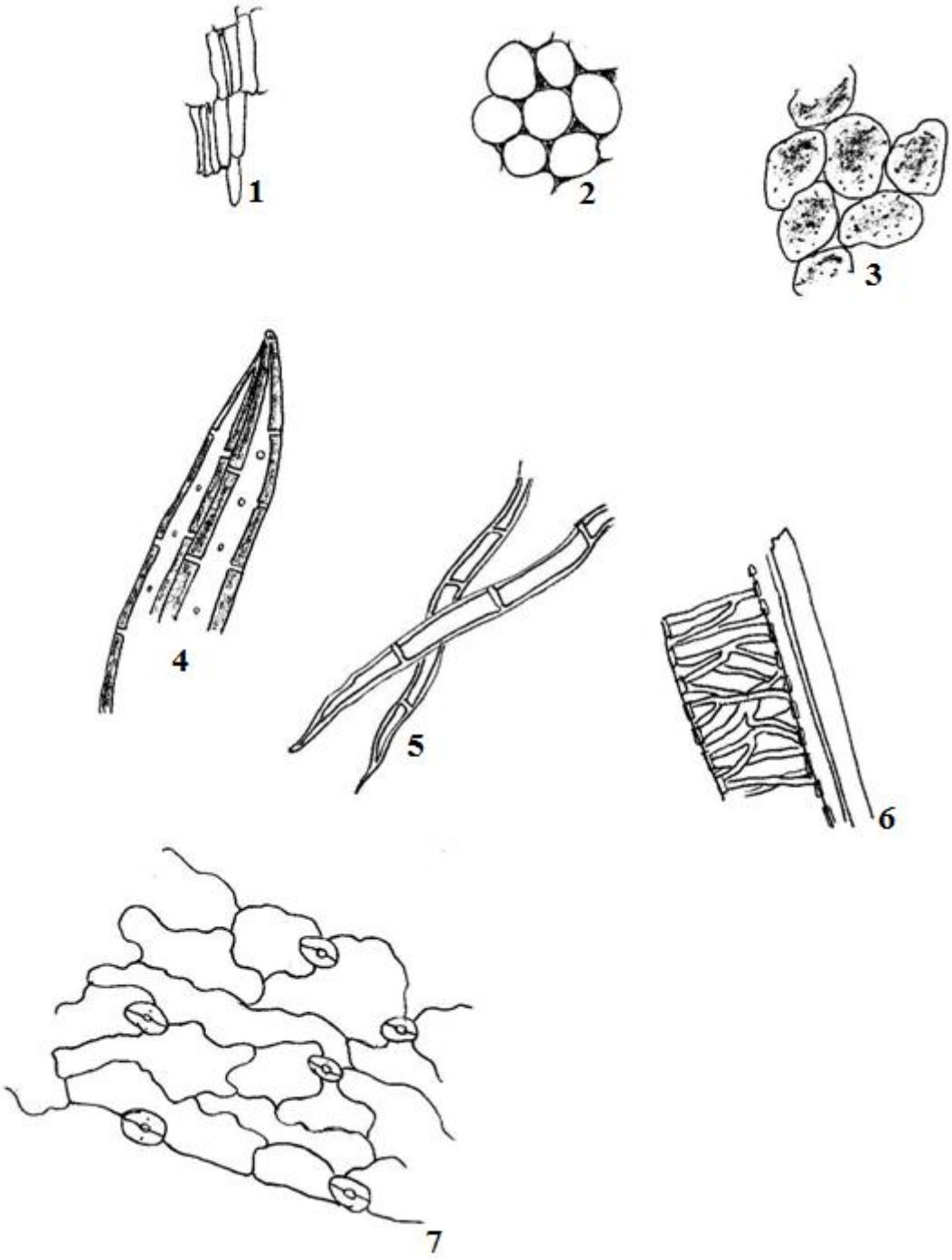

Fig.8. ***Fumaria parviflora*** **powder study**:1. Cork, 2. Collenchyma, 3. Parenchyma with light brown deposits, 4. Sclerenchyma, 5. Septet fibers, 6. Reticulate vessel, 7.Epidermal fragments with stomata.

Physico-chemical analysis:

Table 2 : Values obtained for the proximate analysis.

Sr.No.	Parameter	Mean ± SD (%)*			Average (%)
		Summer	Monsoon	Winter	
1.	Total Ash Content	4.97±0.12	5.16±0.19	5.09±0.22	5.07
2.	Acid Insoluble Ash content	1.01±0.09	1.12±0.11	1.08±0.09	1.07
3.	Alcohol soluble extractives	14.39±0.16	14.32±0.18	14.46±0.11	14.39
4.	Water soluble extractives	16.22±0.42	16.32±0.34	16.39±0.23	16.31

*Each value is a mean of 3 reading

3.b.*Justicia procumbens* Linn.(Acanthaceae)

Synonyms : ***Rostellularia procumbens*** **L. Ness**

Sanskrit : Kavacanamaka, Pansuparyaya, Parpata, Renu, Varatikta.

Vernacular names:

Hindi : Kagner, Makhania Ghas.

Kannada : Nela Bevu, Nucchu Nelabevu.

Malayalam : Tsjeru-Tardavel.

Marathi : Ghatipithpapra, Ghati-Pittapapada.

Tamil : Ottippul, Nerei-Poottie, Kotakacalai, Ampalakkotakam.

Distribution and habitat

The plant is erect or procumbent to ascending herb found throughout India mostly as a weed in moist places.

Morphological features

Stems 30-60 cm high, somewhat woody below, much-branched, subquadrangular. Leaves 2.5-4 by 1-2 cm, ovate-elliptic or elliptic-lanceolate, acute or acuminate, more or less lineolate, glabrous, base usually acute. Flowers in axillary and terminal narrow spikes 3-15 cm long; bracts shorter than the calyx, ovate, acuminate, with scarious margins, minutely scabrous at the tip, otherwise glabrous; bracteoles as long as and similar to the bracts but narrower. Calyx 4-partite nearly to the base; segments with scarious margins, lanceolate, acute, unequal, minutely scabrous at the tip. Corolla upto 0.5 cm long, pale- purple, slightly pubescent outside; upper lip 0.25 cm long, the lower portion ovate, the apical part subquadrate, subtruncate and slightly notched at the apex; lower lip very slightly 3-lobed at the rounded apex. Filaments glabrous except at their insertion. Ovary glabrous; lower part of style pubescent. Capsules long, shortly pointed, oblong, grooved on the back, glabrous. Seeds sub concentrically rugose.

Medicinal uses

The whole plant used in fever, pain due to pharyngolaryngcal swelling and cancer(Chen *et.al*, 1996).In India the decoction of leaf used in asthma (Savithramma *et.al*, 2007)and root in fever due to typhoid (Joshi and Joshi, 2000).

Previous Phytochemical reports

Apigenin, quercetin 7 - O - α - L -rhamnopyranoside , luteolin 7 - O -β - D - glucopyranoside, apigenin7 -O- β- D - glucopyranoside, apigenin 7 - O -

neoperidoside ,β - sitosterol ,β - daucosterol , scopoletin , lupenyl acetate , cycloeucalenol , friedelin , epi- friedelinol , and asiatic acid , luteolin , and quercetin (Zhang, 2006).

Previous pharmacognostic reports

No pharmacognostic work has been done on any part of this plant.

Materials and methods

The plant material has been collected from Vadodara, Gujarat. Phytochemical analysis of the plant for their secondary metabolites such as alkaloids, flavonoids, phenolics, steroids, quinones, mucilages and other compounds were done by standard methods of chromatography and spectrophotometry described in Chapter 2. Pharmacognostic, HPTLC finger printing and physico-chemical studies were done by using standard methods described in Chapter 2.

Results

Phytochemistry

The plant is found to contain flavonoids such as 6-0H kaempferol and 7- OMe 6- OH kaempferol. The phenolic acids were vanillic, syringic and ferulic (*cis*- and *trans*- isomers) acids. Mucilage amounted to 6.8% consisting of rhamnose, glucose and xylose. The plant also showed the presence of unidentified alkaloids and steroids.

Pharmacognosy

Root : T.S (Fig.8.a)

The T.S of the root was circular in outline with a large central woody region. The cork was poorly developed and consisted of 2 to 4 rows of rectangular to slightly tangentially elongated cells and were thick walled. The secondary cortex was very narrow consisting of 5 to 8 rows of thin walled parenchymatous cells. The cells were large polygonal in shape and were

compactly packed. Few of them were found filled with rosette crystals. Endodermis was indistinct. The narrow phloem zone consisted of 6 to 9 rows of cells made up of usual phloem elements. The phloem rays were thin walled and uni- to biseriate. Wood consisted of vessels, tracheids, fibers, parenchyma and rays. Medullary rays were radially elongated and uni- to biseriate with simple pits on their walls. Vessels were broad, simple and bordered pitted occurred singly or in groups. Some of the vessels showed the elongated pits laid parallel. The vessels were more in the centre.

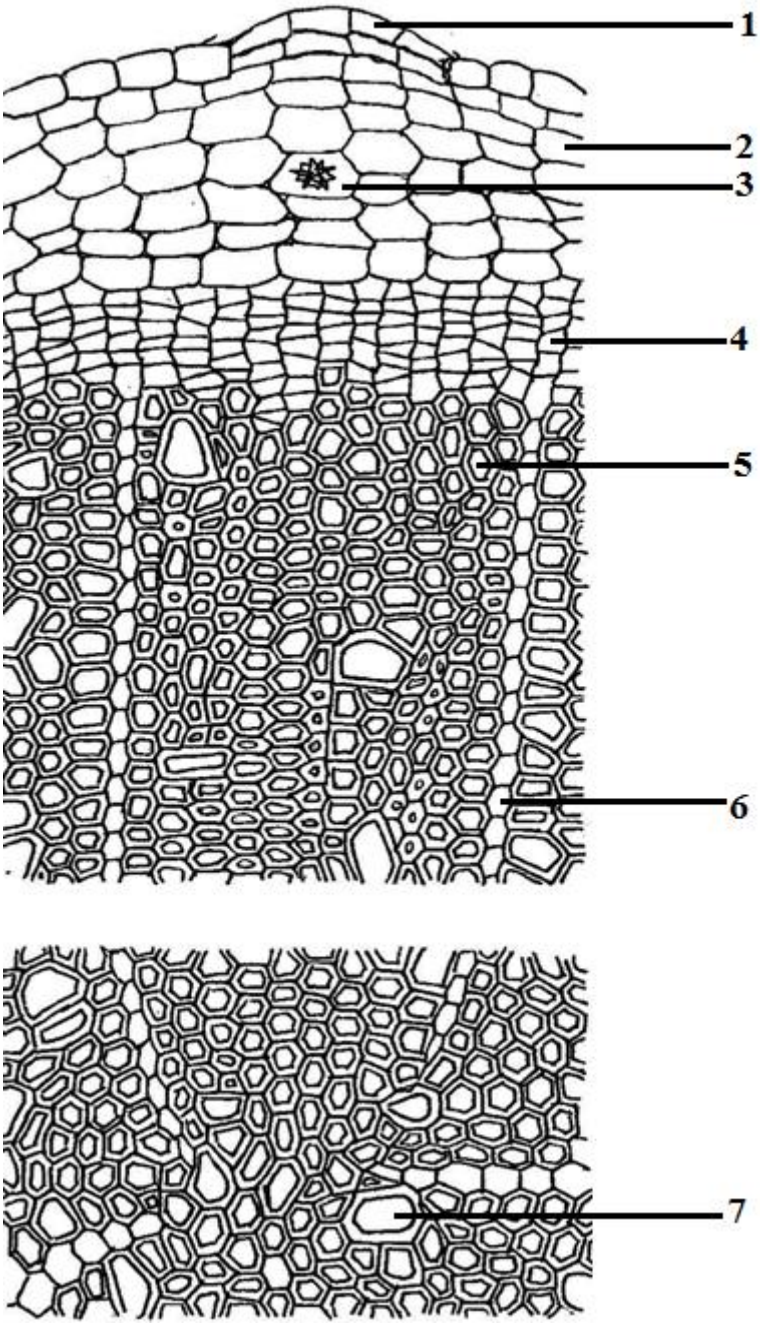

Fig.8.a. ***Justicia procumbens*** **root,T.S**:1.Cork, 2.Cortex, 3.Parenchyma with rosette crystals, 4.Phloemrays, 5.Xylem, 6.Xylem rays, 7.Vessel.

Root : T.L.S (Fig.9)

Cork cells appeared rectangular. The phloem rays were compressed spindle shaped and thin walled and simple pitted. Fibers were thin walled and broad lumened. The xylem rays were spindle shaped with simple pits on their walls. The vessels had 3-4 rows of elongated parallel bordered pits.

Root : R.L.S (Fig.10)

The phloem rays were thin walled. Vessels were broad with bordered pits. The xylem ray cells were appeared rectangular and pitted. The vessels were with alternate boarded pits.

Leaf micromorphology

The stomata were of diacytic type. The stomatal index was 17-19. The trichome index was 9-12. The trichomes were of glandular and non glandular types.The non glandular trichomes were thick-walled unicellular as well as multicellular uniseriate showing broad basal cell, blunt tip and the warty walls. Unicellular trichomes was rare. The glandular trichomes were with a short stalk and circular head made up of two to four cells.

Leaf : T.S (Fig. 11)

The **midrib portion** was characterized by a concave bulge on the upper side and hemispherical bulge on the lower side. The epidermal cells were polygonal in shape covered by thick cuticle. Here the lateral walls of the epidermal cells were characteristically thin and wavy while outer walls were thick and convexly arched outwards. Some of the epidermal cells were circular and showed the deposition of spherical cystoliths. The hypodermis on both upper and lower regions made up of angular collenchyma. The ground tissue was parenchymatous and the
cells on the upper side were rounded and compactly arranged. The vascular bundle was crescent shaped. Below this were large parenchymatous cells. The trichomes were found present on both lower and upper epidermis.

In the **lamina portion (Fig. 12)** the epidermal cells were barrel shaped and the walls were similar to that of midrib. The mesophyll was isobilateral consisted of palisade and spongy tissues. The palisade was single layered and was finely packed with chloroplasts. The spongy tissues contained loosely arranged parenchyma with intercellular spaces.

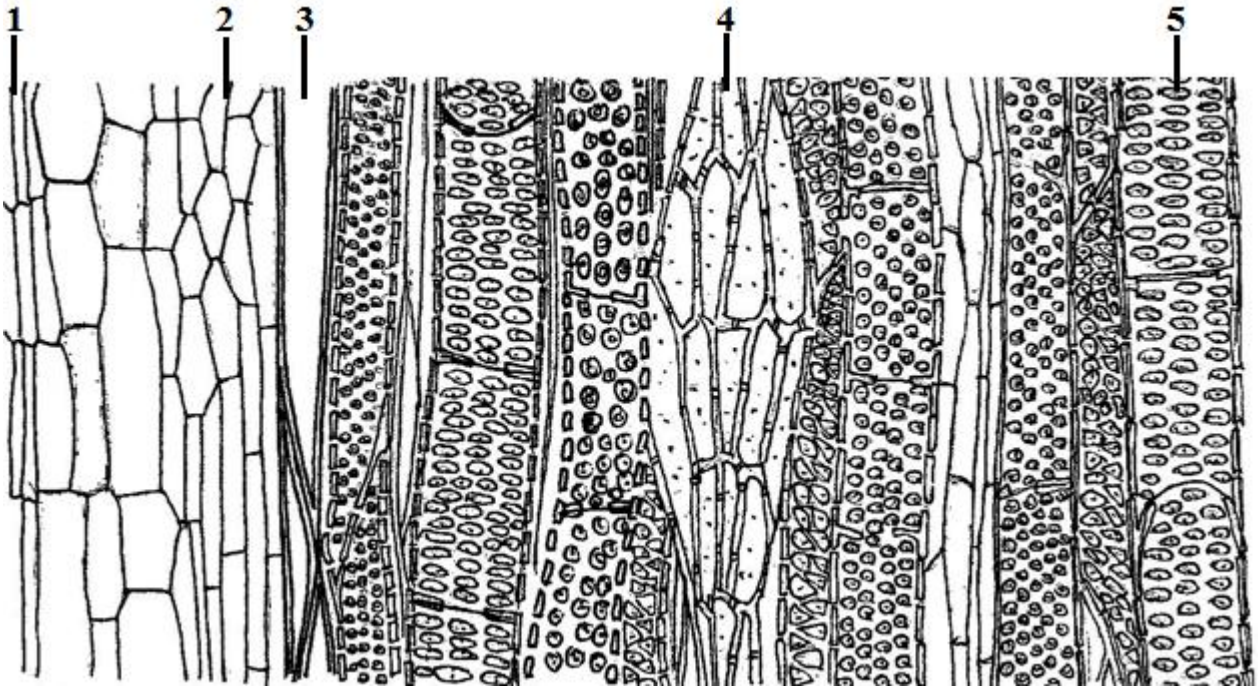

Fig.9.*Justicia procumbens* root, T.L.S:1. Cork cells, 2. Compressed spindle shaped phloem rays , 3. Fibers with broad lumened,4. Spindle shaped xylem rayes, 5. Vessels with elongated parallel bordered pits.

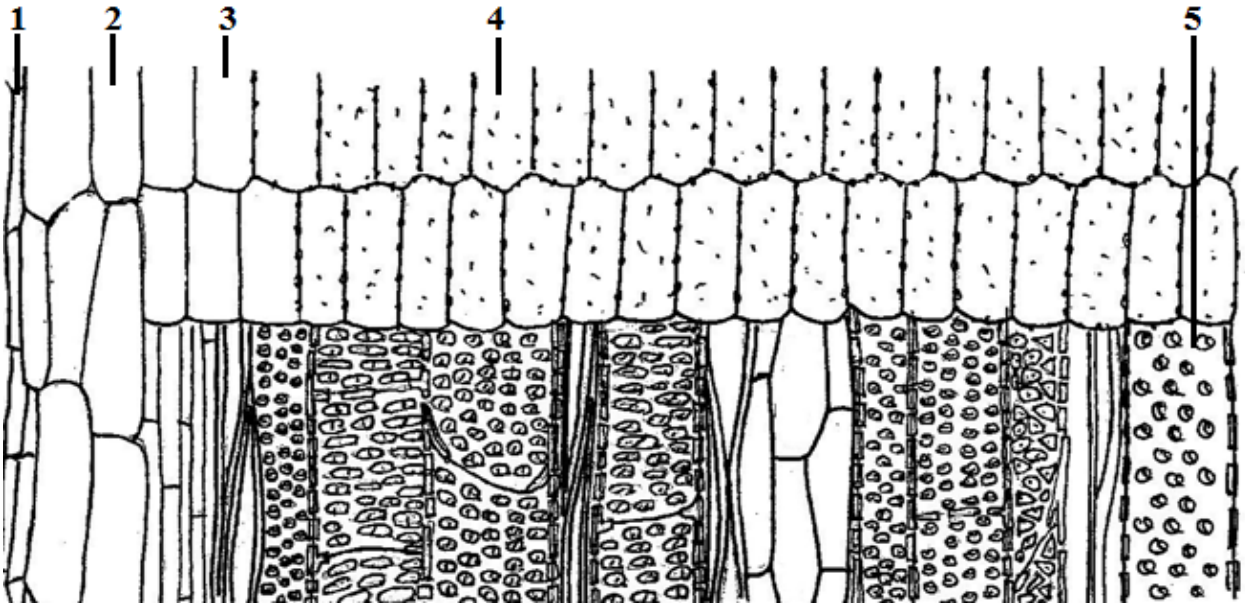

Fig.10. *Justicia procumbens* root, R.L.S:1. Cork cells, 2. Cortex, 3. Phloem rays,4. Xylem rays, 5. Vessels with alternate boarded pits.

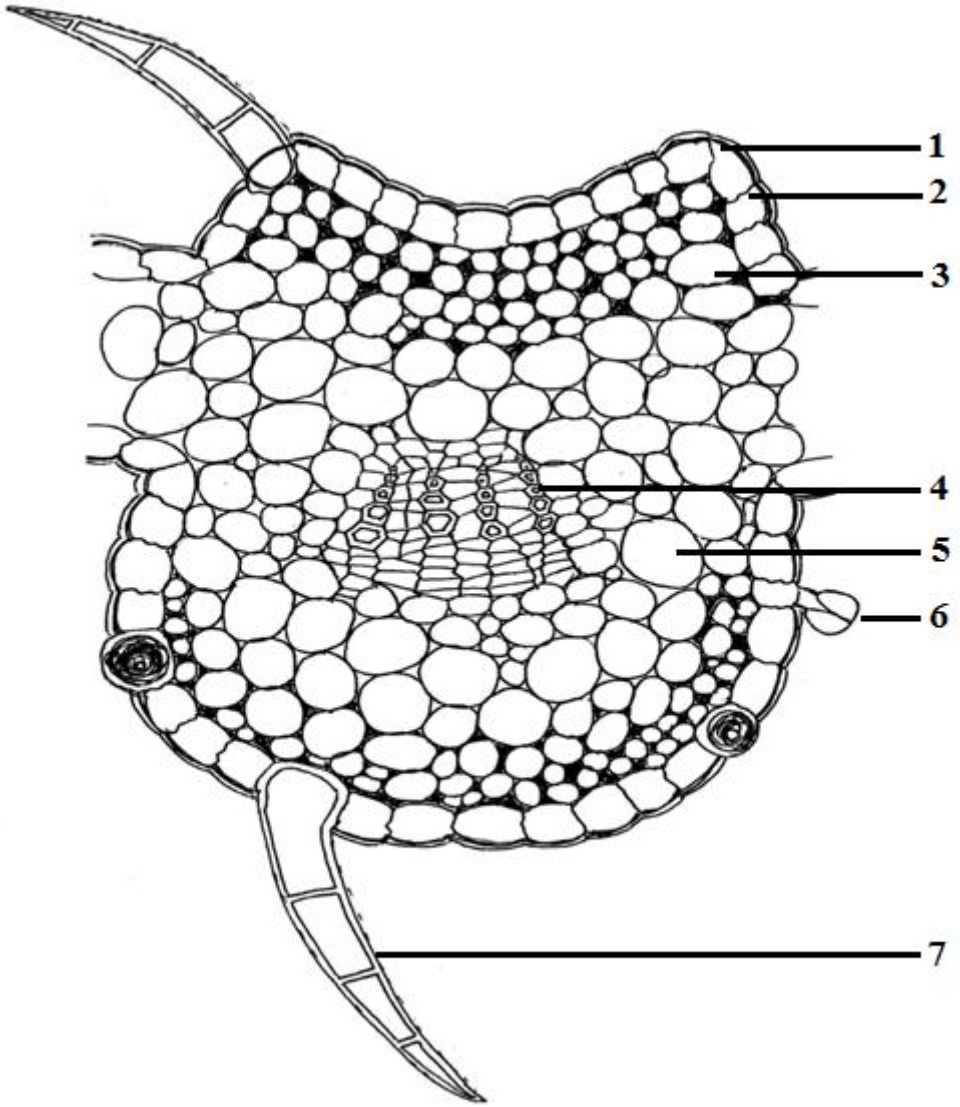

Fig.11. ***Justicia procumbens*** **leaf midrib,T.S**: 1. Epidermis with cuticle, Epidermal cells with wavy lateral walls, 3.Collenchyma, 4. Vascular bundles, 5.Parenchyma, 6. Glandular trichomes, 7. Multicellular trichome,

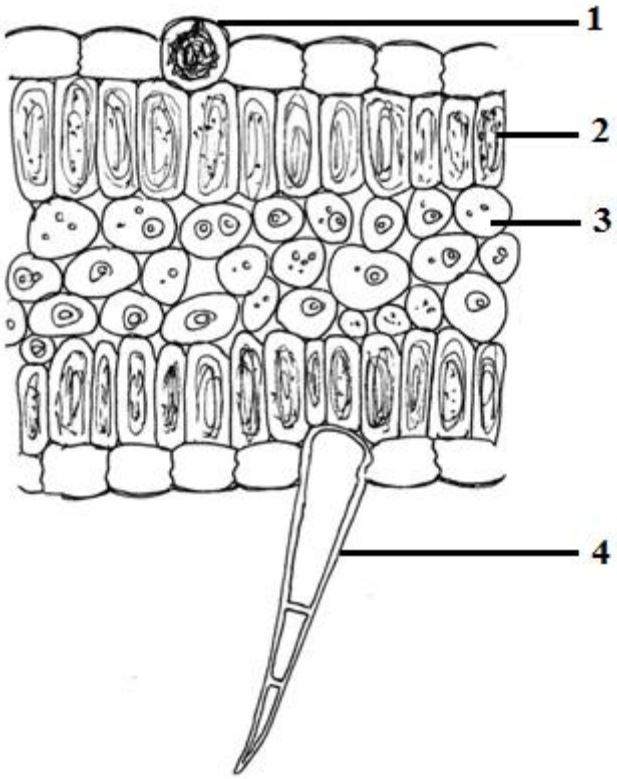

Fig.12. ***Justicia procumbens*** **leaf lamina, T.S**: 1. Epidermis with thick outer walls and spherical cystolith, 2. Palisade cells, 3. Spongy parenchyma, 4. Multicellular trichome.

Stem : T.S (Fig.13)

The T.S. of stem was quadrangular. The epidermis consisted of tabular cells covered with a thin cuticle and contained thick-walled unicellular as well as multicellular uniseriate trichomes with broad basal cells. The tip of the trichomes were blunt and the walls were warty. Some of the epidermal cells became large circular and contained cystoliths. The collenchymatous hypodermis was 3-4 layered developed at the angles of the quadrangular axis. The cortex consisted of 5-7 layers of large polygonal thin walled parenchyma. The endodermis was indistinct. The pericycle was composed of nearly continuous ring of sclereids. The wood composed of large xylem bundles at the angles connected by strands of interfascicular wood prosenchyma with a few rows of vessels embedded in them. The wood parenchyma was occured in groups on the inner side of the angular xylem bundles. The phloem was a narrow zone consisting of usual elements of phloem. Xylem consisted of vessels, tracheids, fibres, parenchyma and xylem rays. Vessels were simple and boarded pitted. Few scalariform vessels were also found.The xylem rays were uniseriate. The central region of pith contained compactly arranged isodiametric thin walled parenchyma.

Stem : T.L.S (Fig.14)

Epidermal cells were rectangular bearing trichomes followed by thick walled collenchyma. The cells of cortex were also polygonal shaped. Xylem rays were pitted and uniseriate. The fibre tracheids were with broad lumen. The pith cells appeared polygonal shaped and thin walled.

Stem : R.L.S (Fig.15)

Epidermal cells were rectangular while the cells bearing cystolith were polygonal in shape. The prosenchyma were also polygonal in shape and thick walled. Xylem rays were rectangular and had pits on their walls.

Powder study (Fig.16)

The components present in the powder were thick-walled unicellular as well as multicellular uniseriate trichomes having broad basal cell and warty walls with blunt tip, glandular hairs, fragments of collenchyma, thick walled prosenchyma, boarded pitted vessels and scalariform vessel.

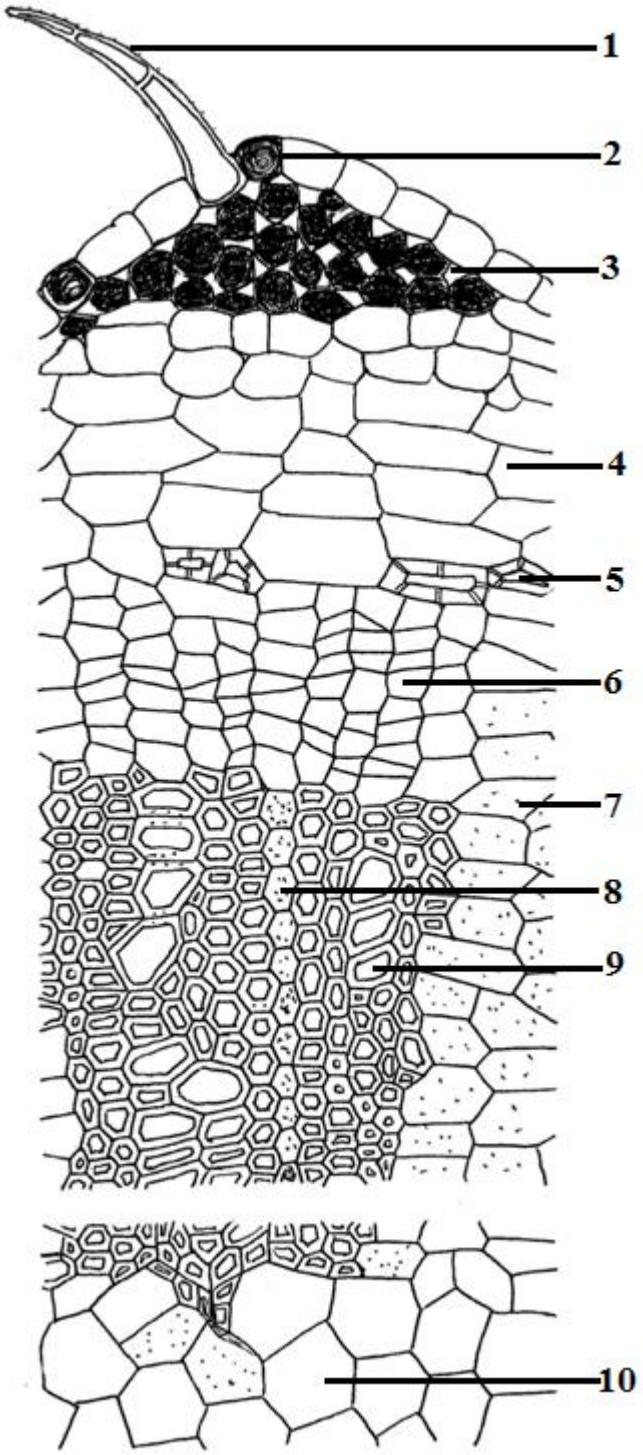

Fig.13. ***Justicia procumbens*** **stem, T.S**: 1. Multicellular uniseriate hair, 2. Epidermal cell with cystolith, 3. Collenchyma, 4.Cortical parenchyma, 4.Sclerenchyma, 6. Phloem, 7. Prosenchyma, 8. xylem rays, 9.Vessel, 10. Pith.

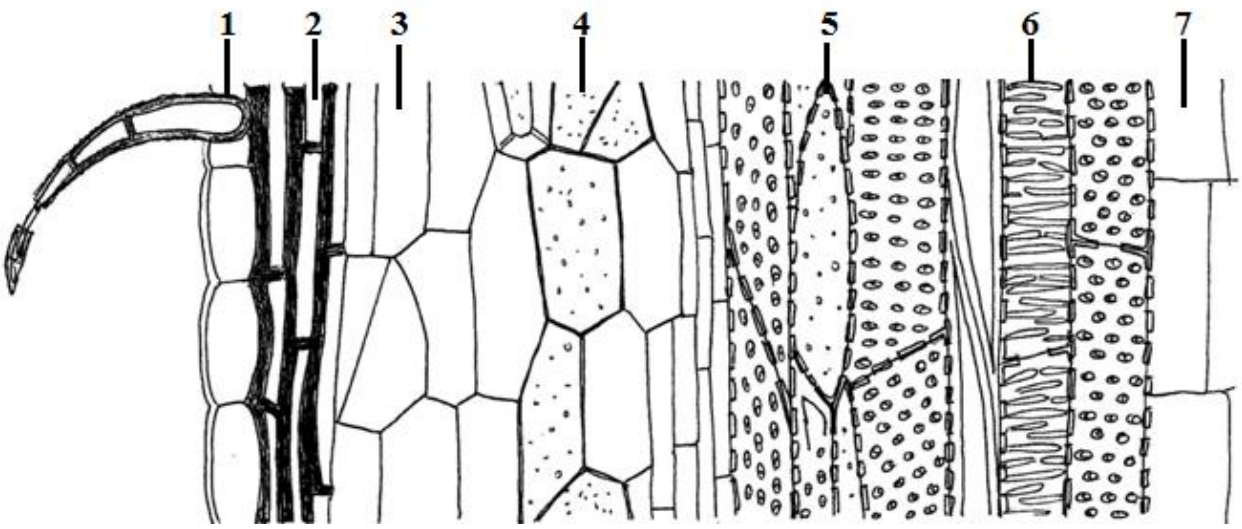

Fig.14. ***Justicia procumbens*** **stem, T.L.S**: 1. Epidermal cell with hair, 2.Collenchyma, 3. Cortex, 4.Prosenchyma, 5. Xylem rays, 6. Scalariform vessel, 7.Pith.

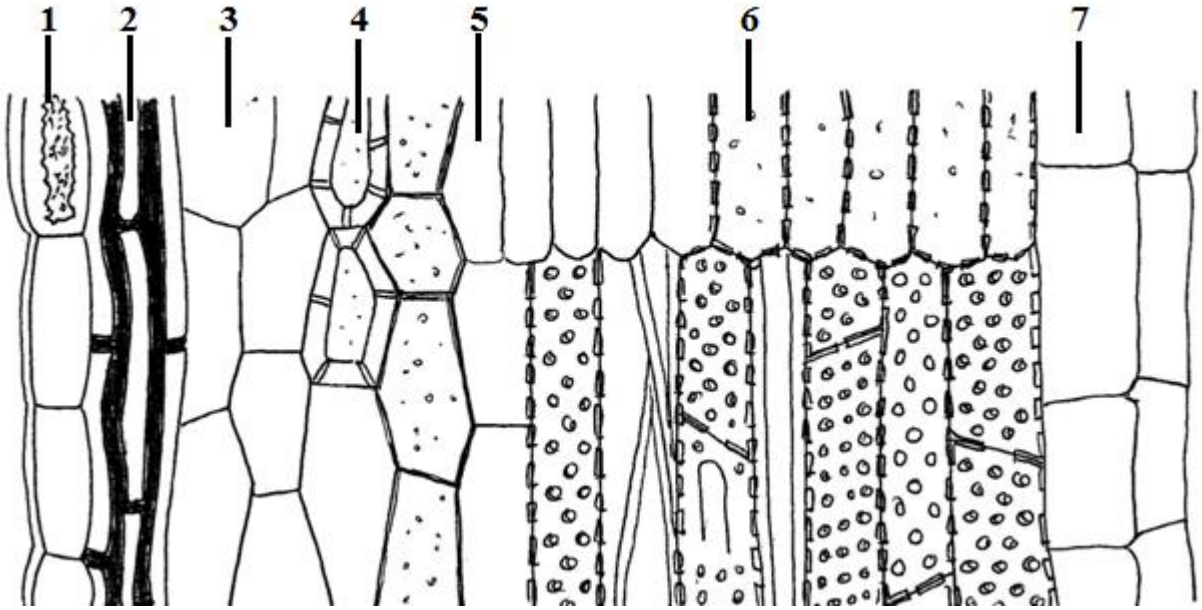

Fig.15. ***Justicia procumbens*** **stem, R.L.S**: 1. Epidermal cell with Cystolith, 2.Collenchyma, 3. Parenchyma, 4. Sclerenchyma, 5. Phloem rays, 6. Xylem rays, 7.Pith.

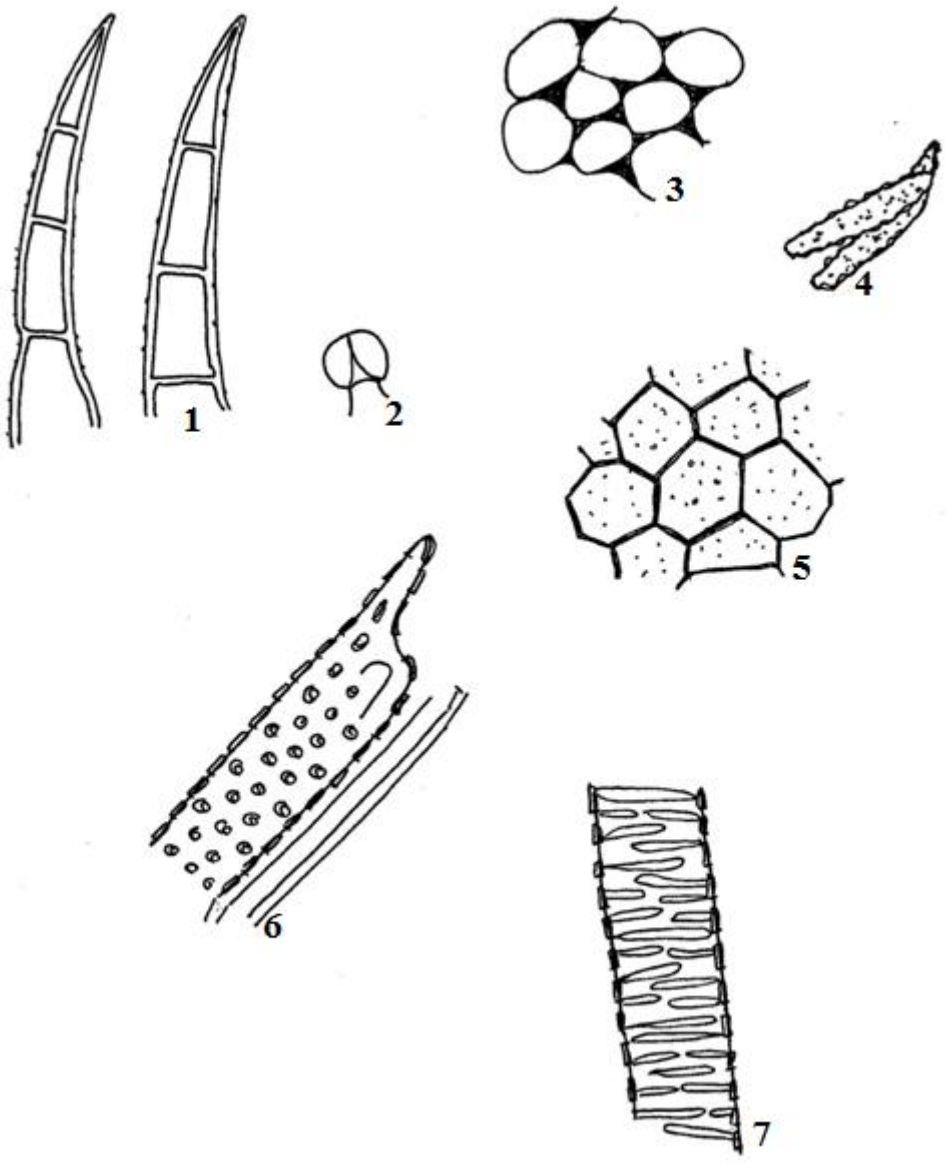

Fig.16. ***Justicia procumbens*** **powder study**: 1. Broad basal celled trichomes with thick, warty walls, 2. Glandular hairs, 3. Fragments of collenchyma, 4. Cystoliths, 5. Fragments of prosenchyma, 6. Boarded pitted vessel, 7. Scalariform vessel.

Distinguishing features

Phytochemical markers

1. 6-OH Kaempferol.
2. 7- OMe 6- OH Kaempferol.
3. Ferulic (*cis*- and *trans*- isomers) acid.

Pharmacognostic markers

Root

1. Centre wood dominated by vessels.
2. Elongated parallel bordered pits.

Leaf

1. Thick-walled unicellular as well as multicellular uniseriate trichomes with broad basal cell, blunt tip and warty walls.
2. Thin and wavy lateral walls of epidermal cells.
3. Thick outer epidermis walls.
4. Diacytic type of stomata.
5. Isobilateral mesophyll.

Stem

1. Thick-walled unicellular as well as multicellular uniseriate trichomes with broad basal cell, blunt tip and warty walls.
2. Interfascicular wood prosenchyma.
3. Broad lumen fibre tracheids.

Physico-chemical analysis:

Table:3. Values obtained for the proximate analysis.

Sr.No.	Parameter	Mean ± SD (%)*			Average (%)
		Summer	Monsoon	Winter	
1.	Total Ash Content	11.02±0.21	11.08±0.17	11.16±0.20	11.09
2.	Acid Insoluble Ash content	0.98±0.03	0.98±0.06	0.99±0.2	0.98
3.	Alcohol soluble extractives	13.33±0.62	12.81±0.66	12.39±0.89	12.84
4.	Water soluble extractives	17.39±0.41	17.22±0.23	17.31±0.28	17.31

*Each value is a mean of 3 reading

3.c. *Oldenlandia corymbosa* L. (Rubiaceae)

Synonyms: *Hedyotis corymbosa* (L.) Lamk

Sanskrit: Ksetraparpatra, Kshetraparpata, Parapataparpata,Parpataka.

Vernacular names:

Bengal : Khetpapra.

Gujarati : Khetpapra , Parpat.

Hind : Daman-Paper, Damanpapar.

Kannada : Kallasabatrasige, Hutcchu Nelabevu, Kallu Sabseege.

Malayalam : Parpatakam , Paropatakapulla.

Marath : Parpat, Papti, Phapti, Parpato, Poripath.

Tamil : Parpadagam, Pappanpuntu, Parpatakam, Kattucayaver, Pappan.

Telugu : Verrinelavemu.

Distribution and habitat

A spreading, suffruticose annual, frequently found especially during monsoon in fields throughout India, Sri Lanka, tropical East Asia to Java and the Phillipines.

Morphological features.

The plant is an annual, height varying from 7.5-38 cm.; stems terete, numerous, slender, erect, ascending or spreading, glabrous or pubescent. Leaves subsessile, 2- 4.5cm. by 1.5-4 mm., linear or linear-lanceolate, acute, often with recurved and frequently scabrous margins; stipules short, membranous, truncate, with a few short bristles. Flowers on the filiform pedicels longer than the calyx, usually 2-3 (rarely 1 or very rarely 4) on the top of a very slender axillary solitary peduncle; bract beneath the pedicels 1, 25-1.5 mm. long, subulate. Calyx 2 mm. long, pubescent; teeth narrowly triangular, about equalling the calyx-tube when in flower. Corolla white, 2.5 mm. long; lobes acute, about 1.25mm. long. Capsules globose or sometimes slightly pyriform, somewhat didymous, the top rather flat and not protruded beyond the calyx, glabrous. Seeds pale brown and angular.

Medicinal uses:

The plant is known to clear heat and toxins, activate blood circulation, promote diuresis and relieve stranguria (urinary obstruction). It is also known to act against tumours of the digestive tract lymphosarcoma and carcinoma of the liver and larynx. It is also active against appendicitis, hepatitis, pneumonia, cholecystesis, urinary infection, cellulites and snake bite. Chinese folk medicine describes the plant to treat

skin sores, ulcers, sore throat, bronchitis, gynaecologic infections and pelvic inflammatory diseases (Chang 1986; Bensky 1990;Chang 1992 ;Ming 1990).It is usually administered in the form of a decoction in remittent fever with gastric irritability and nervous depression caused by deranged bile. It is also used to treat jaundice and diseases of liver. The juice of the plant is applied to palms and soles to relieve the burning sensation during fevers. The plant is used as an anthelmintic. In Philippines the plant is boiled in water and the brew is used as a mouthwash for relief during toothache(Anonymous). The methanolic extracts of the plant is found to be antioxidant, radical scavenging, anti-inflammatory, cytotoxic and antibacterial (Nordin and Ahmad 2006). Immunomodulatory studies (Sutarjadi *et.al.*,1991)and hepatoprotective studies (Sadashivan *et.al.,* 2006) have been conducted using the plant extracts.

Previous Phytochemical reports

Iridoids such as geniposide, 6α-hydroxygeniposide, scandoside methyl ester, asperulosidic acid, deacetylasperuloside, asperuloside, 10-*O*-benzoylscandoside methyl ester, 10-*O*-*p*-hydroxybenzoylscandoside methyl ester, (+)-lyoniresinol-3α-O-β-glucopyranoside (Tagaki *et al.*,1981; Nishihama *et al.*,1981) and rutin (Noiarsa *et al.*,2008) have been identified. Acylated derivatives such as 10-O-benzoyl deacetyl asperulosidic acid methyl ester, and 10-O-benzoyl, 10-O-*p*-hydroxybenzoyl, and the 10-O-*p*-*trans*, *cis*-coumaroyl scandoside methyl esters have also been isolated and identified (Hideaki *et al.*,1991). The plant is also known to contain oleanolic and ursolic acids (Nordin and Ahmad. 2006). The plant is also known to contain alkaloids biflorine and biflorone (Nordin and Ahmad. 2006).

Previous pharmacognostic reports

No study has been done on the pharmacognostic characters of the plant.

Materials and methods

The plant material has been collected from Vadodara, Gujarat.Phytochemical analysis of the plant for their secondary metabolites such as alkaloids, flavonoids, phenolics, steroids, quinones, mucilages and other compounds were done by standard methods of chromatography and spectrophotometry described in Chapter 2. Pharmacognostic, HPTLC finger printing and physico-chemical studies were done by using standard methods described in Chapter 2.

Results

Phytochemistry

The plant is found to contain flavonoids such as quercetin, 3'-methoxy quercetin and 3', 4'-dimethoxy quercetin. The plant contained the anthocyanin cyanidin and pelargonidin, while the phenolic acids located were vanillic, syringic, *p*-coumaric (*cis* and *trans* isomers), *p*-hydroxybenzoic, gentisic and caffeic acids. Mucilage amounted to 7.6% consisting of glucose and xylose. The plant also showed the presence of unidentified alkaloids and steroids.

Pharmacognosy

Root : T.S. (Fig.17)

The root was circular in outline. Cork was a narrow zone made up of 2-4 layers of thin walled cells. Cortex was of 5-9 layers of somewhat broadly rectangular parenchymatous cells, many of them contained bundles of raphides. Phloem was 10 to 12 layers thick and made up of usual elements. The wood composed of small vessels, fibre tracheids, parenchyma and rays with the patches of libriform fibres. The libriform fibres were thick walled, narrow lumened and having blunt ends. Xylem rays were uniseriate with few biseriate and having pits on their walls. The vessels were comparatively small.

Root : T.L.S (Fig.18)

The cork cells were many layered and rectangular. Cortical parenchyma contained bundles of raphides. Phloem rays were thin walled and appeared spindle shaped. Libriform fibre were found associated with tracheids. Xylem vessels had alternate simple pits.

Root : R.L.S. (Fig.19)

Cortical parenchyma were square to rectangular shaped and contained raphide bundles. The Phloem rays were thin walled. The xylem rays consisted of rectangular shaped pitted cells. Vessels were bordered pitted.

Leaf micromorphology

The stomata were of paracytic type. The stomatal index was 21-25. The leaf was free from trichomes or glands.

Leaf : T.S (Fig.20)

The **midrib portion** was characterized by a concave cavity on the upper side and a hemispherical bulge on the lower side.The epidermal cells were polygonal in shape covered by thin cuticle. The hypodermis on both upper and lower regions made

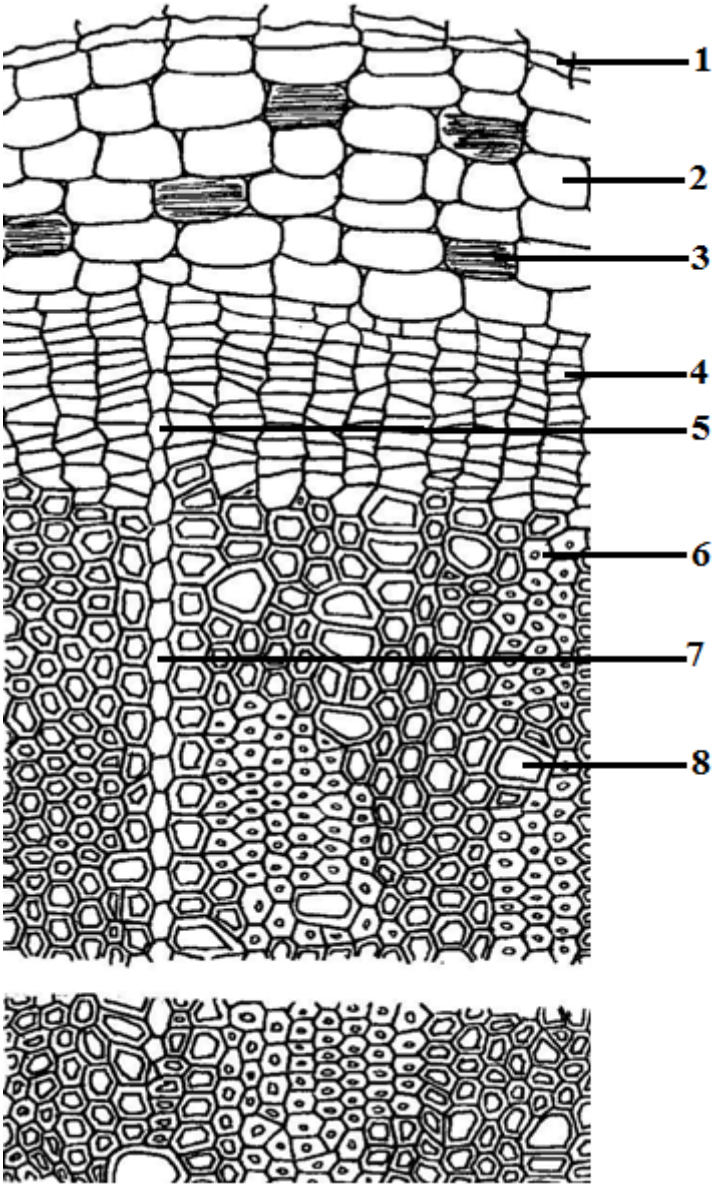

Fig.17.*Oldenlandia corymbosa* root, T.S: 1. Cork, 2. Cortex, 3. Parenchyma with raphides bundles, 4. Phloem, 5. Phloem ray, 6. Libriform fibres, 7. Xylem rays, 8.Vessel.

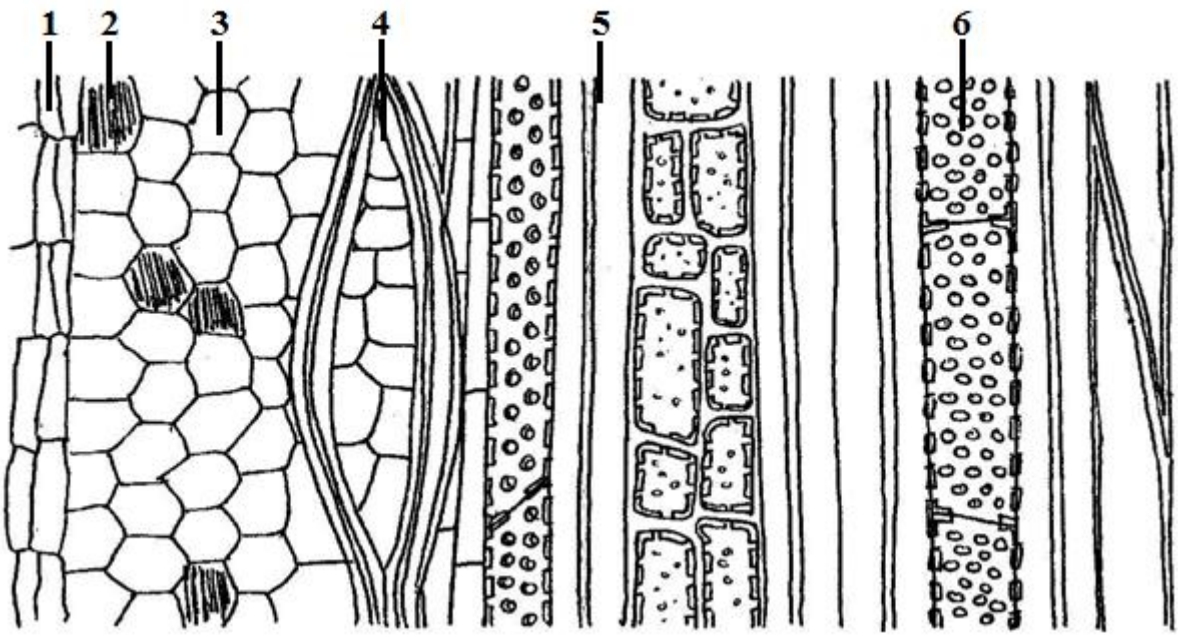

Fig.18. ***Oldenlandia corymbosa*** **root, T.L.S**:1. Cork, 2. Raphides bundles, 3. Cortex, 4. Spindle shaped ray, 5. Libriform fibres, 6.Vessel.

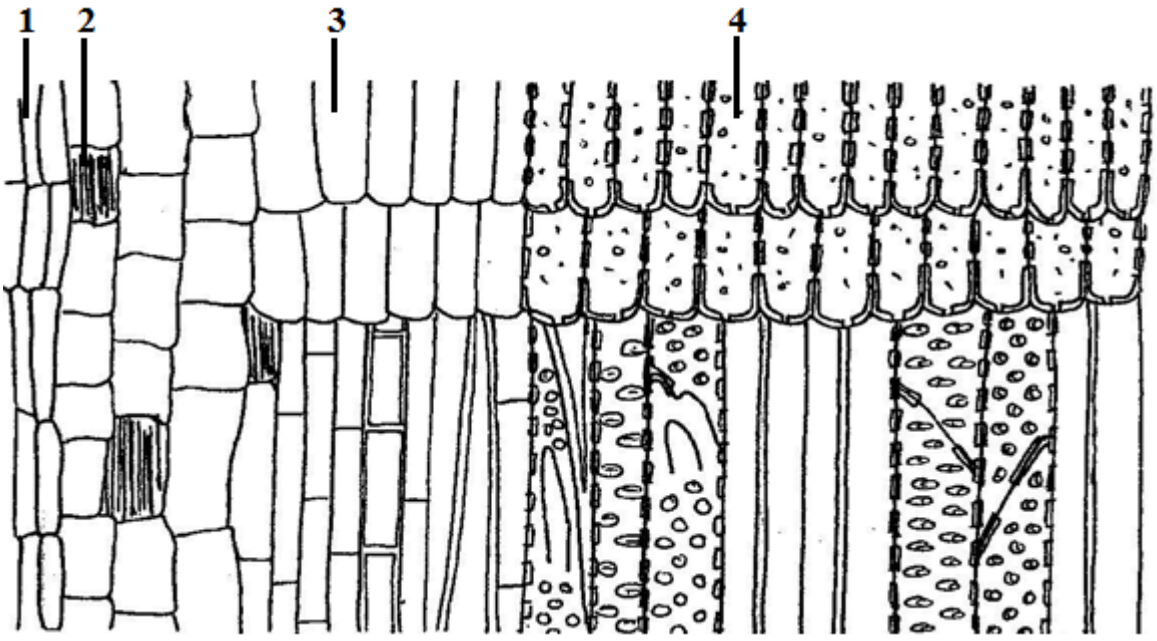

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Fig.19. ***Oldenlandia corymbosa*** **root, R.L.S**:1. Cork cells, 2. Raphides bundles, 3.Phloem rays, 4. Xylem rays.

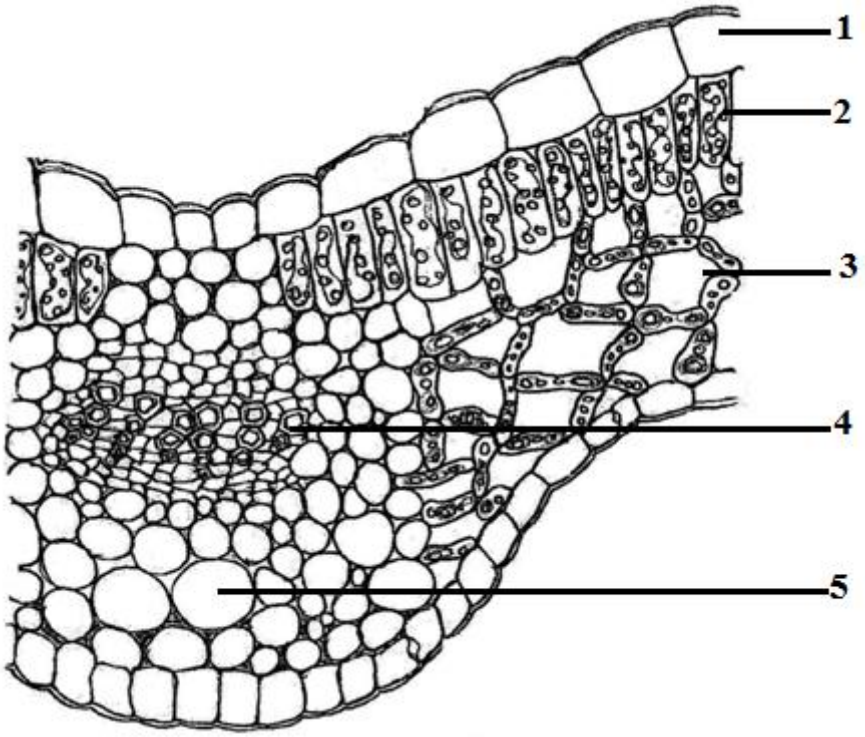


Fig.20.*Oldenlandia corymbosa* leaf, T.S: 1. Epidermis with cuticle, 2. Single layered palisade, 3. Mesophyll parenchyma, 4. Vascular bundle, 5. Ground tissue.

up of collenchyma. The ground tissue was parenchymatous. The cells of the ground tissues on the upper side of the vascular bundle were smaller and spherical while those on the lower side were composed of large isodiametric parenchyma cells.The vascular bundle was crescent shaped.

In the **lamina portion** the epidermal cells were barrel shaped. The cells of the upper epidermis were about double the size of the lower epidermis. The mesophyll cells were differentiated into a single layer of palisade and lower spongy tissue. The palisade cells were filled with large chloroplasts. The mesophyll region was almost double the size of the palisade, consisting of a network of lobed spongy cells containing chloroplasts. Air spaces within were very large. The leaf was free from trichomes or glands.

Stem : T.S (Fig.21)

The T.S. of stem was almost square in outline. The epidermis consisted of barrel shaped cells. At some places there were cork formation in the outermost layers made up of 3 to 5 layers of thin walled rectangular cells.Cortex was 6 to 8 layers thick and composed of thin walled parenchymatous cells. The cells were circular in shape and compactly packed with very little intercellular spaces. Some of these cells contained raphide bundles and chlorophyll. Endodermis and pericycle were indistinct. Phloem was narrow zone consisting of 5 to 8 layers of cells. The wood was diffuse porous and composed of small vessels, fibres, tracheids, parenchyma and rays. In the xylem the libriform fibres found associated with tracheids. Xylem rays were uni- to biseriate, thick walled and did not contain any inclusions. The vessels were comparatively small and were simple and boarded pitted. The scalariform vessels were also common. The pith in the centre was large and was composed of thin walled large parenchymatous cells.

Stem : T.L.S (Fig.22)

Epidermal cells were thin and rectangular shaped followed by few layers of small square to pentagonal thin walled parenchymatous cells. Some large rectangular cortical parenchyma were found with raphide bundles. The phloem cells were thin rectangular. Xylem rays were pitted and biseriate. The scalariform vessels showed the straight end walls. The pith cells appeared rectangular and were thin walled.

Stem : R.L.S (Fig.23)

Cork cells were thin walled and rectangular in shape. Cortical parenchyma were with raphide bundles. Phloem and Xylem rays were rectangular in shape. Xylem rays showed the pits on their walls.

Powder study (Fig.24)

The colour of the powder were light green and the components present in the powder were fragments of epidermis with stomata, thin walled cortical parenchyma containing raphide bundles, raphide bundles (made up of needle shaped crystals), mesophyll containg chlorenchyma, narrow lumened fiber.

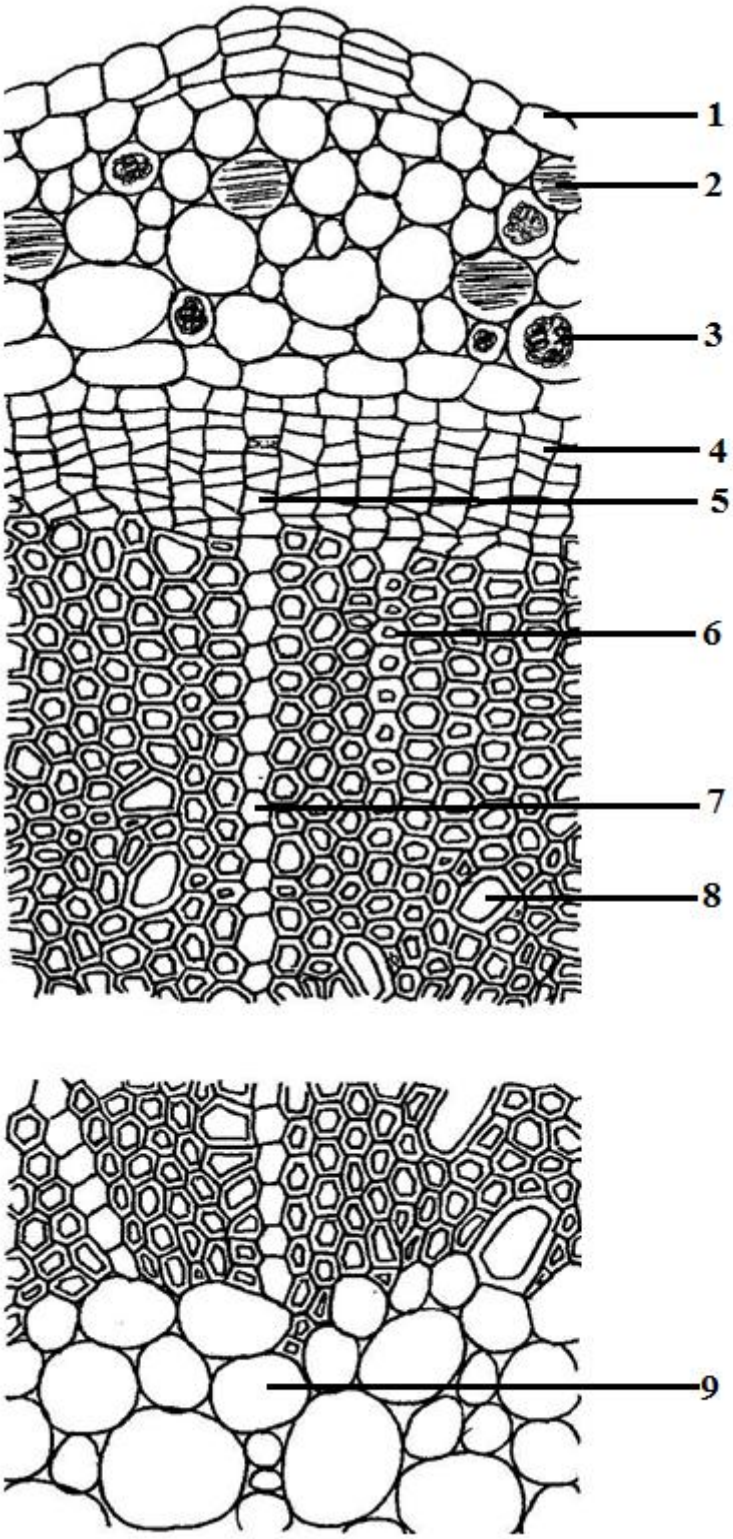


Fig.21.*Oldenlandia corymbosa* stem, T.S: 1. Epidermis, 2. Parenchyma with raphides bundles, 3. Parenchyma with chlorophyll, 4. Phloem, 5. Phloem ray, 6.Libriform fibres, 7. Xylem rays, 8.Vessel, 9.Pith.

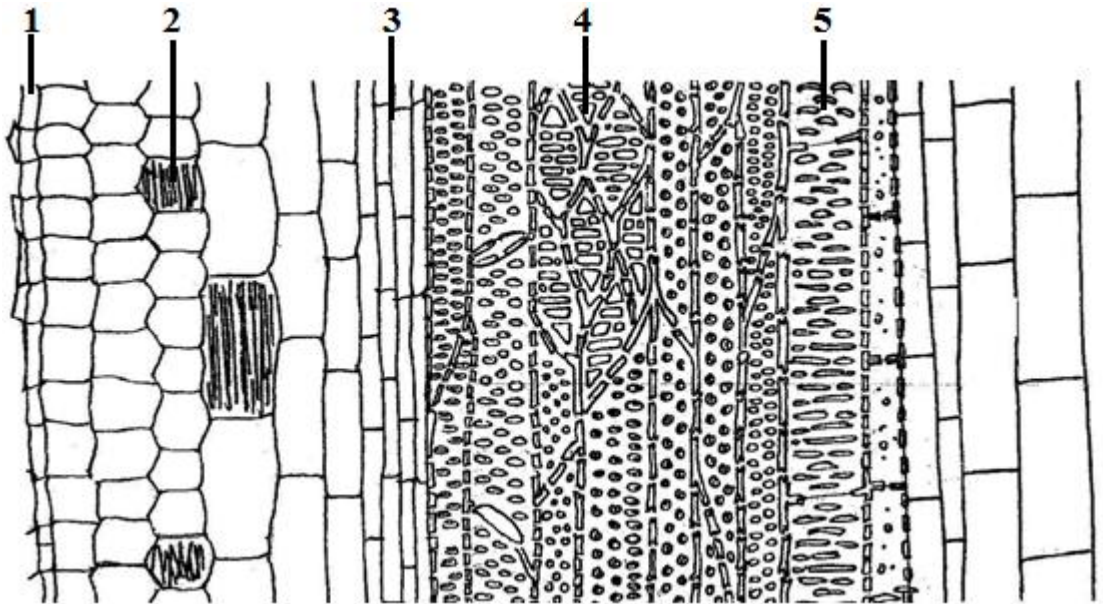


Fig.22. ***Oldenlandia corymbosa*** **stem, T.L.S**: 1. Epidermis, 2.Cortical parenchyma with raphides bundles 3. Phloem, 4. Xylem rays, 5. Scalariform vessel.

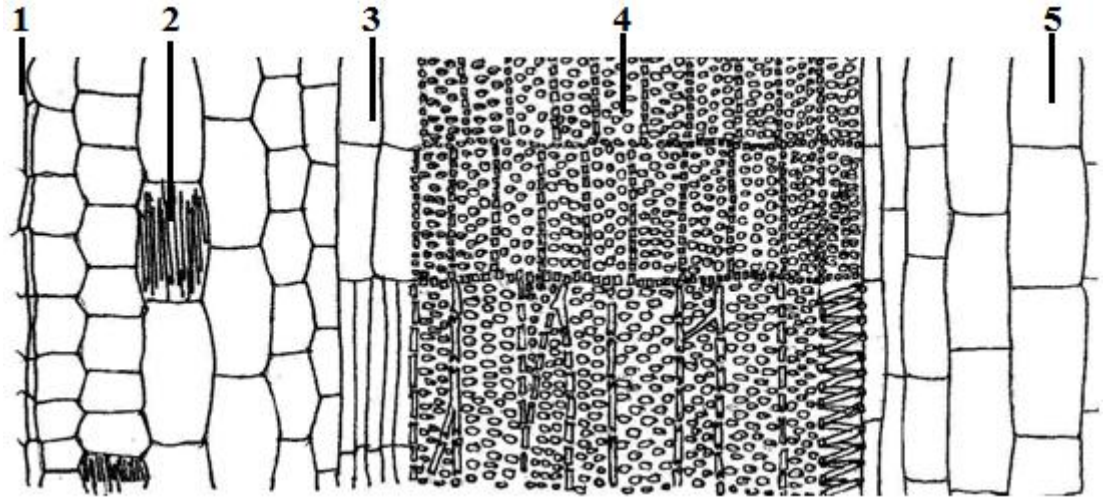


Fig.23. ***Oldenlandia corymbosa*** **stem, R.L.S**: 1. Cork, 2. Raphides bundles, 3.Phloem rays, 4. Pitted xylem rays, 5. Pith.

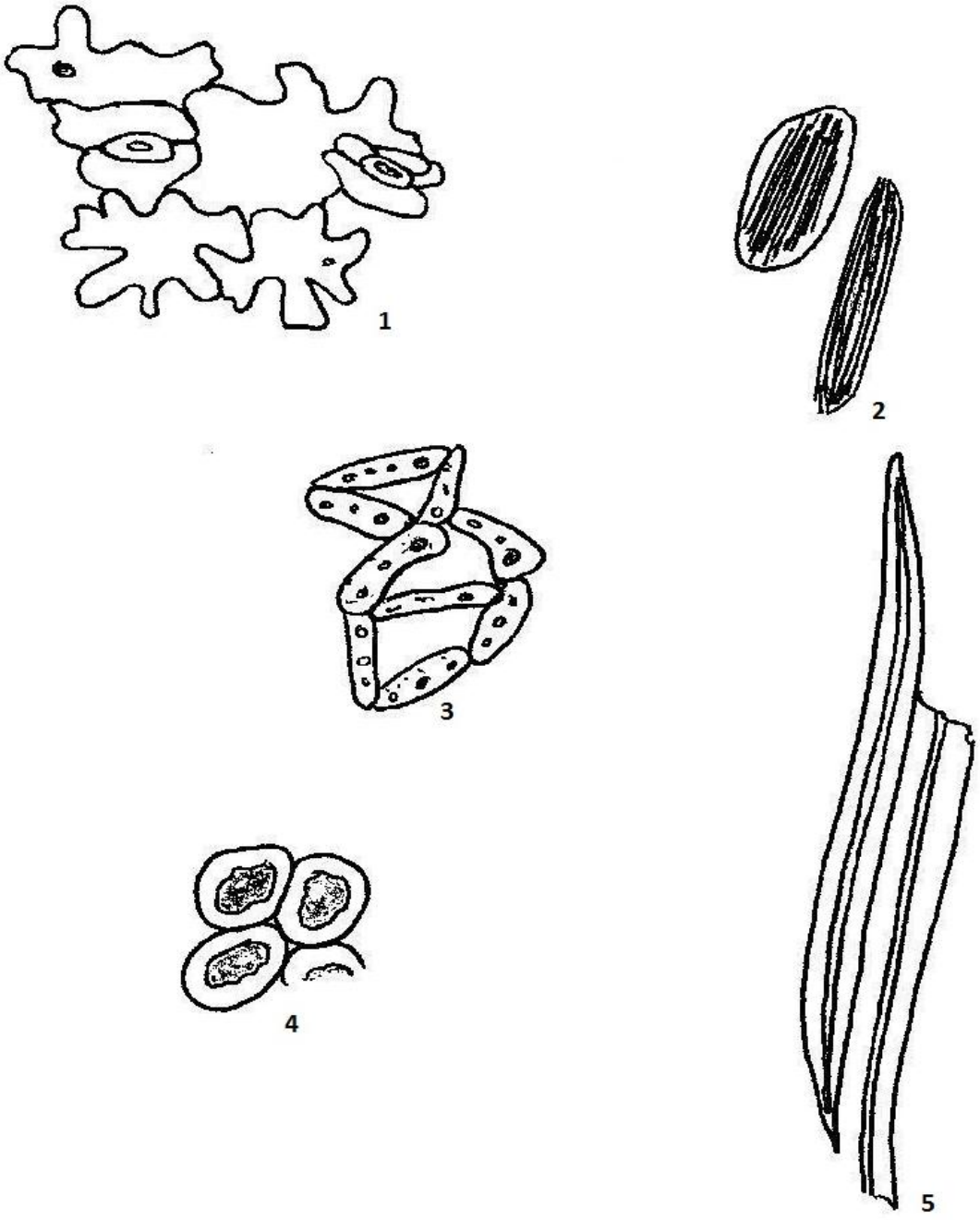


Fig.24.*Oldenlandia corymbosa* Powder study:1. Fragments of epidermis with stomata, 2.Raphide bundles, 3.Mesophyll containing chloroplast, 4.Chlorenchyma , 5. Thick walled narrow lumened fibers.

Distinguishing features

Phytochemical markers

1. 3'-Methoxy quercetin.
2. 3', 4'-Dimethoxy quercetin.
3. *p*-Coumaric (*cis* and *trans* isomers) acid.
4. *p*-Hydroxybenzoic acid.
5. Gentisic acid.
6. Caffeic acid.
7. Cyanidin.
8. Pelargonidin.

Pharmacognostic markers

Root

1. Parenchyma cells bearing raphide bundles .
2. Narrow lumened libriform fibres.

Leaf

1. Cells of upper epidermis about double the size of the lower epidermis.
2. Paracytic type of stomata.
3. Network of lobed spongy cells.
4. Absence of trichomes.

Stem

1. Association of libriform fibres with tracheids.
2. Scalariform vessels with straight end walls.

Physico-chemical analysis:

Table: 4. Values obtained for the proximate analysis.

Sr.No.	Parameter	Mean ± SD (%)*			Average (%)
		Summer	Monsoon	Winter	
1.	Total Ash Content	10.01±0.18	10.11±0.13	10.09±0.14	10.07
2.	Acid Insoluble Ash content	1.18±0.14	1.14±0.12	1.09±0.21	1.14
3.	Alcohol soluble extractives	7.91±0.54	7.26±0.66	7.22±0.46	7.46
4.	Water soluble extractives	15.31±0.27	15.11±0.09	15.13±0.11	15.18

*Each value is a mean of 3 reading

3.d.*Peristrophe bicalyculata* Nees. (Acanthaceae)

Synonyms: *Peristrophe paniculata* (Forssk.) Brummitt

Sanskrit: Kakajangha, Nadikanta, Sulomasha.

Vernacular names:

Gujarati : Kaliadhedi, Kariadhedi, Lasiadhedi.

Hindi : Kali aghedi ,Atrilal, Kakajangha.

Manipuri : Khuan langthrei.

Marathi : Ghatipittapapada, Ramkiayat, Pitpapra.

Tamil : Nagananda, Chebisa.

Malayalam : Katou-pulcholli.

Telugu : Chebura, Chebira.

Kannada : Cheebee gida, Cheebera Soppu.

Bengali : Nasabhaga.

Distribution and habitat

The plant is an erect hispid herb or under shrub 60-180 cm high, found throughout in India in forest as undergrowth, hedges and wasteland.

Morphological features

The plant is herbaceous, 1-1.5 m high; stems and branches usually 6-angled, more or less hairy, usually rough on the angles. Leaves 5-8 cm long, ovate, acuminate, densely linolate, more or less hairy above, somewhat densely so on the nerves and veins beneath, base usually rounded; main nerves 4-6 pairs; petioles upto 1 cm long. Flowers in trichotomous cymes in large lax divaricate pubescent panicles; bracts beneath the calyx 2, opposite, often 1 cm long, linear, acute, mucronate, with white membranous margins; bracteoles 4, similar to the bracts but shorter, subequal or sometimes unequal. Calyx divided to the base; segments lanceolate-subulate with ciliolate margins. Corolla rosy, nearly 1.5 cm long, pubescent outside; bilabiate upper lip elliptic-oblong, obtuse, entire; lower lip slightly longer, oblong, with 3 acute lobes. Filaments hairy; anther-cells one almost entirely about the other, muticous. Ovary pubescent at the tip; style nearly glabrous. Capsules 1 by 0.4 cm, narrowed into a long, pointed, pubescent cylindric stalk. Seeds orbicular, papillose and slightly rugose.

Action and uses:

In ethnomedicinal practices the traditional healers use the plant as an antidote to snake poison and in bone fractures and sprains (Anon.1966).The ethanolic extract of the plant has been reported to exhibit analgesic, anti-inflammatory and antibacterial properties (Chopra,1959 and Dwivedi, 2002) and was strongly effective against *Staphylococcus aureus*, *Klebsiella* spp., *E. coli*, and *Pseudomonas aeruginosa* (Giwa *et al*,2010).The plant also showed the blood pressure lowering and hepatoprotective effects (Abdulazeez *et al*, 2010).

Previous Phytochemical reports

The chemical composition of the dried aerial parts showed 14-methyl-tritriacont-14-en-15-ol and 35-hydroxynonatriacontanal (Singh *et. al.,* 2000). The volatile oil contained beta-caryophyllene (33.9%), alpha-zingiberene (10.4%), germacrene D and globulol (5.0%) were the compounds occurring in abundance and phytol (56.3%), 1, 8-cineole (20.4%), with sizeable proportions of alpha-pinene (7.1%) and p-cymene (4.0%) (Ogunwande *et. al.*, 2010).

Previous pharmacognostic reports

Very little data available on pharmacognosy of this plant. Only the T.S of various parts of the plant has been studied (Anon.2001 and Saraswathy *et.al.*, 2006).

Materials and methods

The plant material has been collected from Vadodara, Gujarat. Phytochemical analysis of the plant for their secondary metabolites such as alkaloids, flavonoids, phenolics, steroids, quinones, mucilages and other compounds were done by standard methods of chromatography and spectrophotometry described in Chapter 2. Pharmacognostic, HPTLC finger printing and physico-chemical studies were done by using standard methods described in Chapter 2.

Results

Phytochemistry

The plant contained alkaloids in traces and flavonoids were found absent. But it was rich in phenolic acids such as vanillic, syringic, ferulic, coumaric (*cis*- and *trans*-isomers), caffeic and p-hydroxy benzoic acids. Mucilage amounted to 5.8% consisting of xylose. The plant also showed the presence of coumarins.

Pharmacognosy

Root : T.S (Fig.25)

The root was circular in outline in T.S. The cork cells were broad, thin walled and rectangular. The cortex made up of 4 to 8 layers of tangentially elongated polygonal parenchymatous cells. The cells were slightly thick walled and were compactly arranged and few of them were filled with the simple round starch grains (11-21µm). Endodermis was single layered and the cells were barrel shaped and slightly thick walled. Phloem was 6 to 9 layered and composed of phloem parenchyma, fibers and sieve elements. Phloem rays were indistinct. Xylem was dominated by trachieds and fibers. The vessels were scanty and mostly occurred singly in the periphery and in a groups of 2-3 in the center. Xylem rays were thin walled tangentially elongated, uni- to biseriate.

Root : T.L.S (Fig.26)

Cortical cells appeared rectangular and some of the cells here were filled with starch grains. The phloem rays were small spindle shaped. The fibres were straight with a narrow lumen. The vessels were very broad and contained 3-5 rows of transversely oblique bordered pits. Xylem rays were uniseriate to biseriate and contained simple pits.

Root : R.L.S (Fig.27)

Cork cells appeared small rectangular. A single vertical row of thick walled endodermis was seen. Phloem rays were thin walled and without pits, while xylem rays were with simple pits. Vessels were reticulate and border pitted.

Leaf micromorphology

Stomata was of diacytic type. Stomatal index was 16-23. Trichomes were multicellular uniseriate with a trichome index of 5-9 and glandular trichomes were very rare.

Leaf : T.S (Fig. 28)

In the **midrib portion** the epidermal cells were barrel shaped except the cells at upperridges which were polygonal in shape. Hypodermal lamellar collenchyma was discontinuously arranged and present only in the ridged portion of upper and lower midrib. Below this was a parenchymatous ground tissue. The cells of the ground tissue was loosely arranged. Some of them contained chlorophyll and few

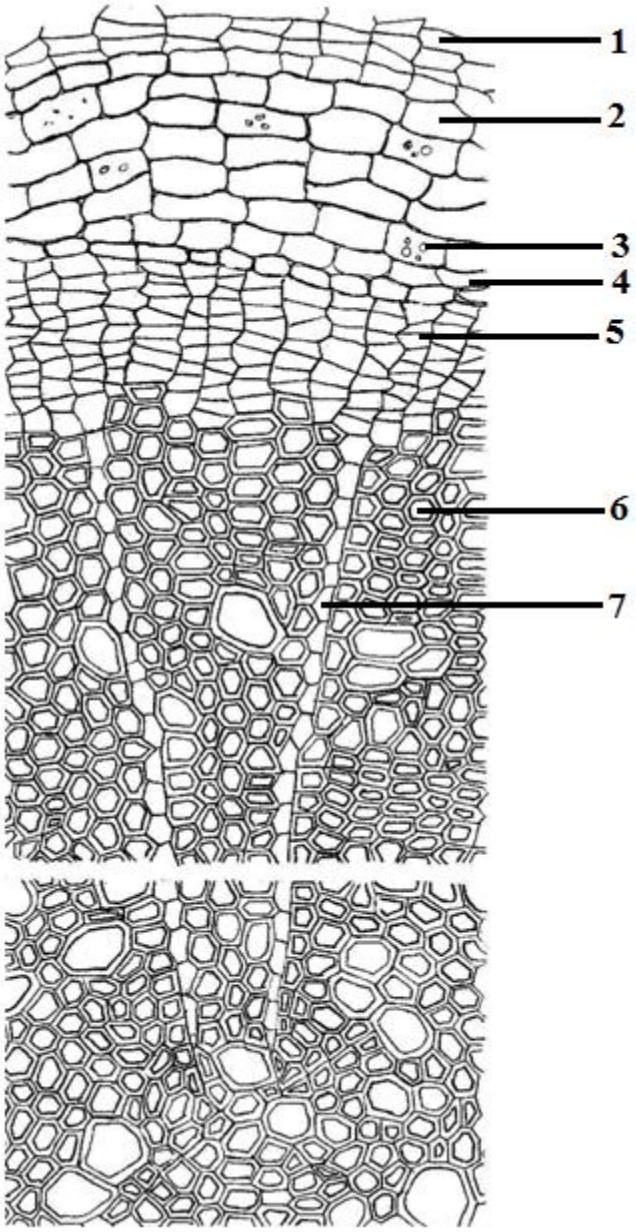


Fig.25. ***Peristrophe bicalyculata*** **root, T.S**.:1. Cork, 2. Cortex, 3.Starch grains, 4.Endodermis, 5.Pholem, 6. Xylem, 7.Xylem rays.

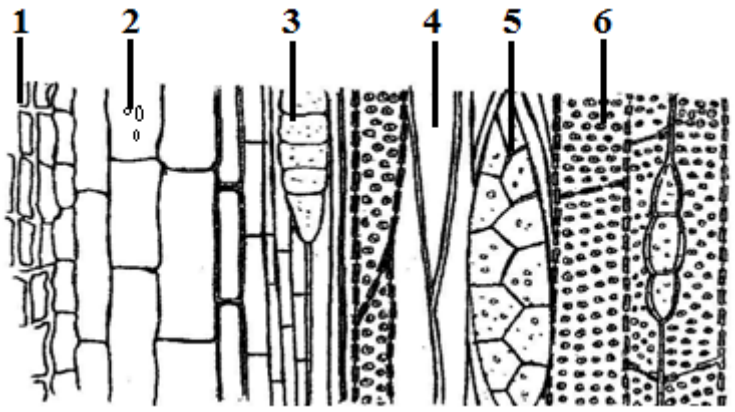


Fig.26. ***Peristrophe bicalyculata*** **root, T.L.S.:**1.Cork, 2. Cortical parenchyma with starch grains, 3. Pholem ray, 4.Fibres, 5. Xylem rays, 6. Vessel.

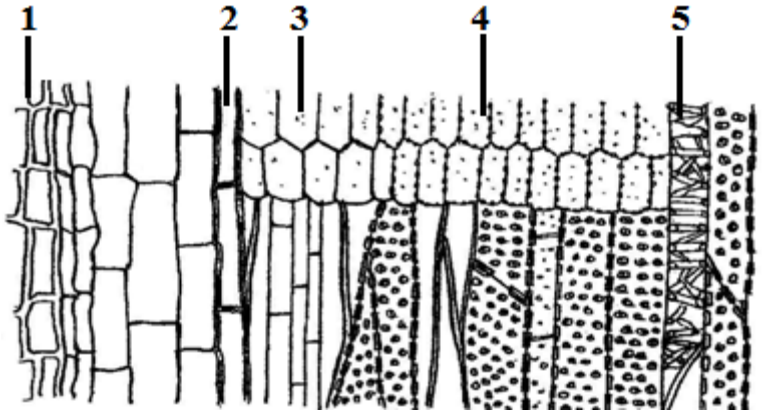


Fig.27. ***Peristrophe bicalyculata*** **root, R.L.S.:**1.Cork, 2. Endodermis, 3. Pholem rays, 4. Xylem rays, 5. Reticulate vessel.

showed the deposition of oil globules. The vascular bundle was crescent shaped. The tracheids were in about 4-6 rows, and there were 3-7 rows of secondary phloem cells. The upper and lower epidermis were covered by thin walled unicellular as well as multicellular uniseriate trichomes with pointed tip and also showed the presence of cystoliths. The 3 celled trichomes were most common. The cells of lower epidermis was smaller than the upper epidermis and also contained a few sessile or short stalked glandular trichomes.

In the **lamina portion** mesophyll was differentiated to an upper single layered palisade and lower 2-3 layered spongy tissue. The palisade cells contained a single row of chloroplasts forming a ring and the spongy cells were with 5-8 large chloroplasts. Trichomes were more on the lower epidermis whereas stomata and cystoliths were seen both on the upper and lower epidermis. Walls of lower epidermal cells were wavy.

Stem : T.S (Fig.29)

The stem in T.S was angular in outline. The single layered epidermis consisted of barrel shaped cells, which was covered by a thin cuticle. Some of the cells contained cystoliths of calcium carbonate. The hypodermis was of 2 to 3 layers of continuous ring consisted of lamellar collenchyma which were 5 to 8 layers thick below the angular protrusion. Cortex was differentiated into outer 1 to 2 layers of chlorenchyma followed by thin walled loosely arranged rounded or oval parenchyma. Some cells here contained brown deposits. Endodermis was single layered of closely fitted thin wall parenchymatous cells some of these cells were became thick walled. Phloem was narrow and composed of phloem parenchyma, phloem fibres and sieve elements. Xylem consisted of vessels, tracheids, xylem parenchyma and fibres. Vessels were scattered and a few of the vessels were in a 3 to 4 rows surrounded by the trachieds. Wide centre pith was parenchymatous. The cells were compactly arranged showing presence of isolated acicular crystals.

Stem : T.L.S (Fig.30)

Epidermal cells contained cystolith. The hypodermal collenchyma were ractangular in shape. Cortical cells were hexagonal in shape. Fibres had uniform lumen sizes. Xylem rays were fusiform 3- cell thick and 5-7 cells in height. In vessels the bordered pits were transversely elongated.

Stem : R.L.S (Fig.31)

In the R.L.S the cortical parenchyma where large polygonal. The phloem and xylem ray cells appeared erect and were of rectangular in shape. The protoxylem had both annular and spiral type of thickenings. Cells of the pith were thin walled, filled with acicular crystals.

Powder study (Fig.32)

The colour of the powder was yellowish green and the components present in the powder were unicellular as well as multicellular uniseriate trichomes having sharp pointed tips, glandular trichomes, epidermal cell containing cystoliths, acicular fibers, vessels, rod shaped crystals.

Distinguishing features

Pharmacognostic markers

Root

1) Cork cells were broad and thin walled.
2) Thick walled prominent endodermis.
3) Simple rounded starch grains.
4) Transversely oblique bordered pits.

Leaf

1) Epidermis were covered by thin walled unicellular as well as multicellular uniseriate trichomes with pointed tip.
2) Deposition of cystoliths in the epidermis.
3) The stomata were of diacytic type.
4) Presence of glandular trichome.
5) Lower epidermal cells were wavy.

Stem

1) cortical cells were showed brown deposits.
2) Pith was parenchymatous showed the presence of isolated acicular crystals.
3) Vessels were surrounded by the trachieds.

Phytochemical markers

1. Ferulic acids.
2. Coumaric (*cis*- and *trans*-isomers) acids.

3. Caffeic acids.
4. *p*-Hydroxy benzoic acids.
5. Xylose.
6. Absence of flavonoids.

Physico-chemical analysis:

Table:5. Values obtained for the proximate analysis.

Sr.No.	Parameter	Mean ± SD (%)*			Average (%)
		Summer	Monsoon	Winter	
1.	Total Ash Content	09.88±0.23	10.09±0.09	09.93±0.21	9.97
2.	Acid Insoluble Ash content	0.53±0.06	0.57±0.03	0.54±0.3	0.55
3.	Alcohol soluble extractive	08.87±0.94	8.88±0.89	8.82±0.91	8.86
4.	Water soluble extractive	11.12±0.63	11.32±0.38	11.22±0.26	11.22

*Each value is a mean of 3 reading

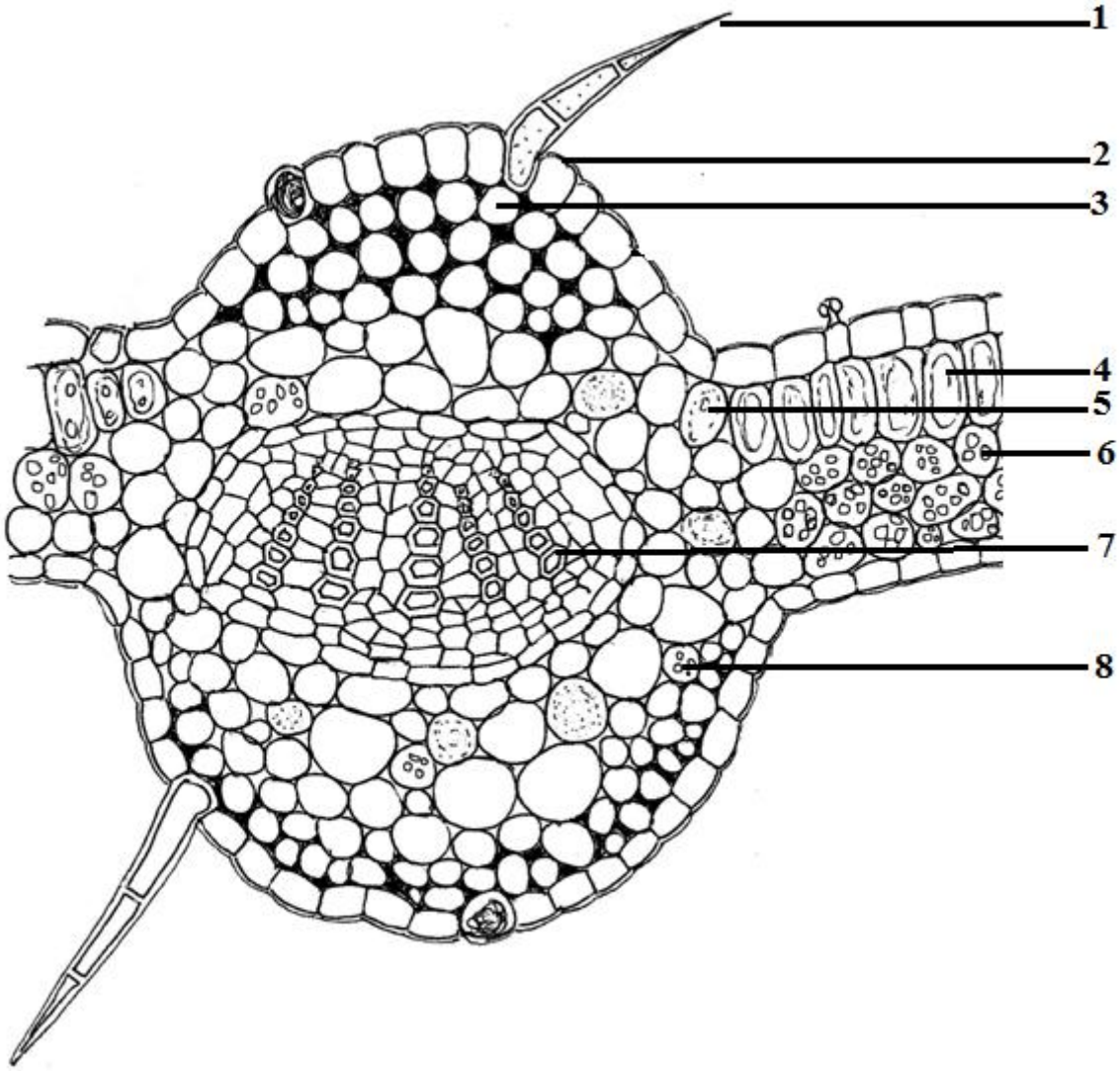


Fig.28. ***Peristrophe bicalyculata*** **leaf, T.S:** 1. Multicellular trichome, 2. Epidermis with cuticle, 3. collenchyma, 4. Palisade, 5.Chlorenchyma, 6. Spongy parenchyma, 7. Xylem, 8. Parenchyma with deposition of globules.

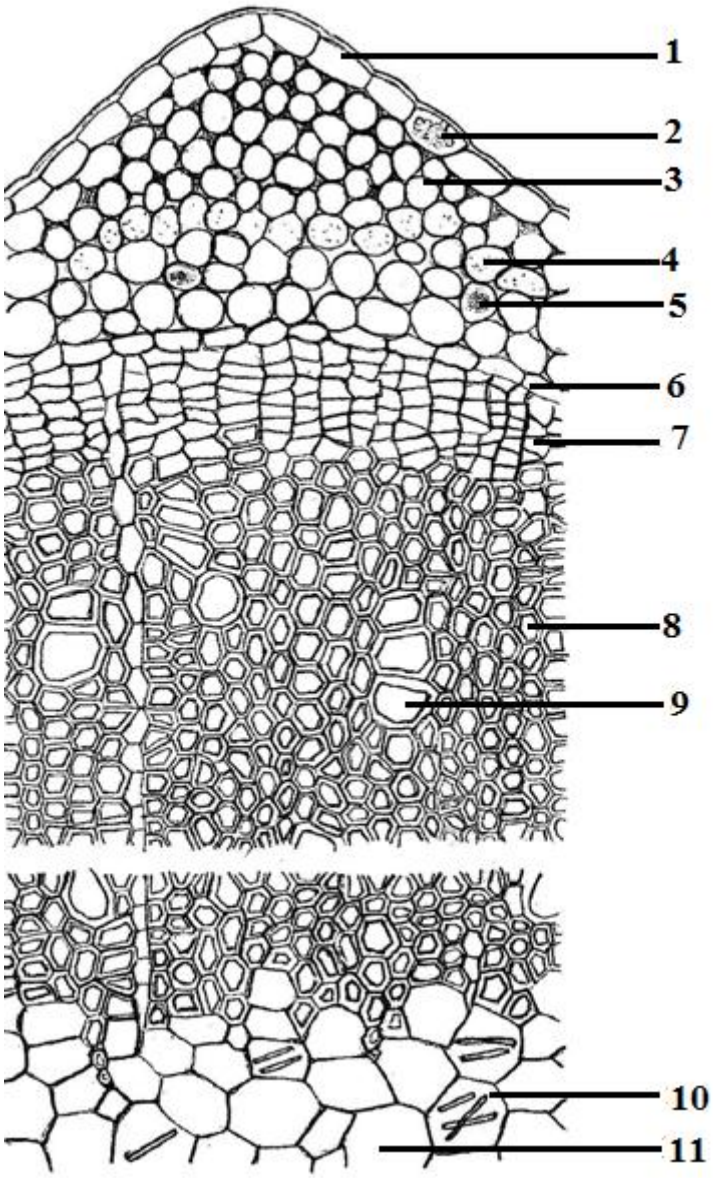


Fig.29. *Peristrophe bicalyculata* stem, T.S: 1. Epidermal cell with cuticle, 2.Cystolith, 3.Collenchyma, 4.Chlorenchyma, 5.Parenchyma with brown deposits, 6. Endodermis, 7. Phloem, 8. Xylem, 9. Vessel, 10. Acicular crystals, 11.Pith.

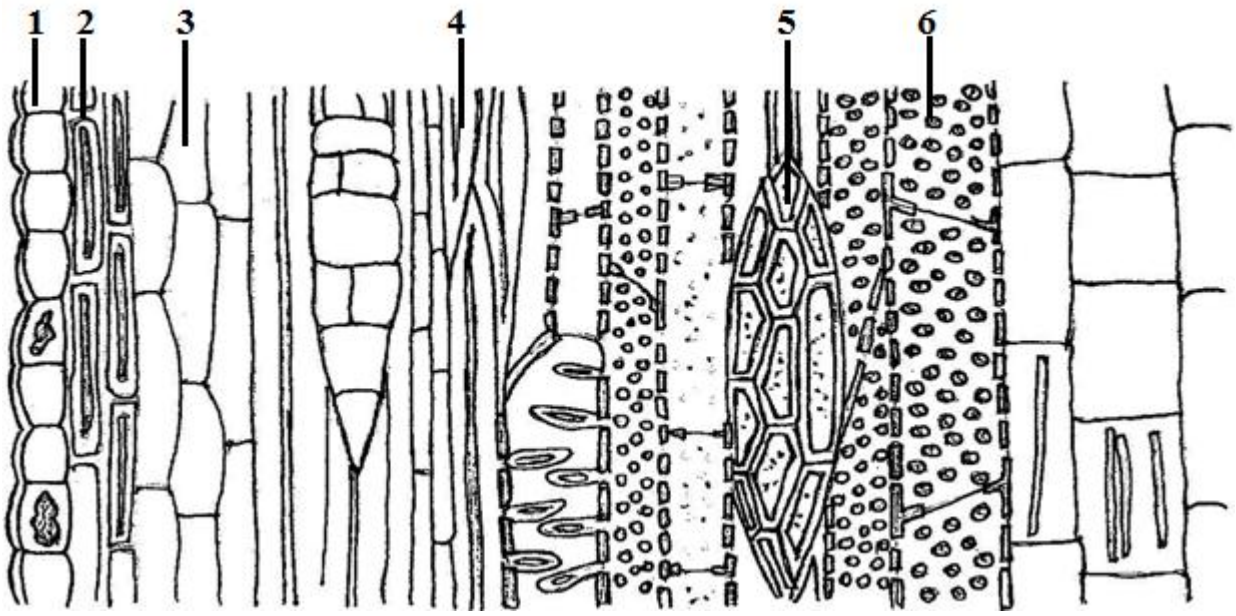


Fig.30. ***Peristrophe bicalyculata*** **stem, T.L.S**: 1. Epidermal cell cystolith, 2. Collenchyma, 3. Cortex, 4. Fibres, 5. . Xylem rayes, 6. Vessel.

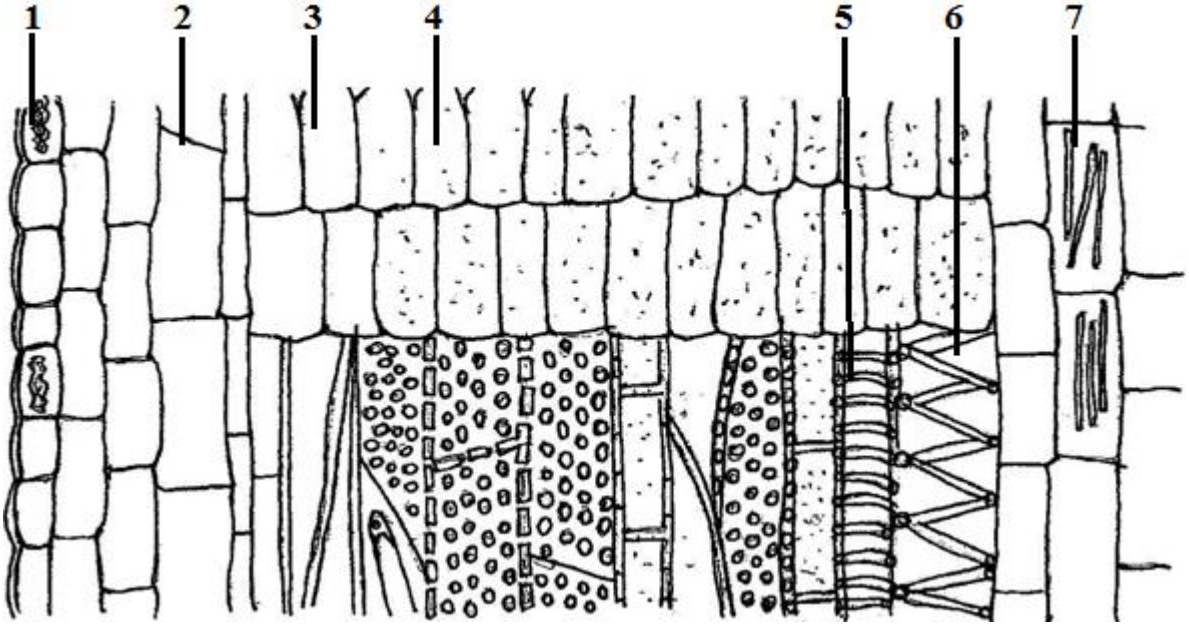


Fig.31. ***Peristrophe bicalyculata*** **stem, R.L.S**: 1. Epidermal cell with cystolith, 2. Cortex, 3.Phloem rays, 4. Xylem rays, 5. Annular vessel, 6. Spiral vessel, 7.Pith.

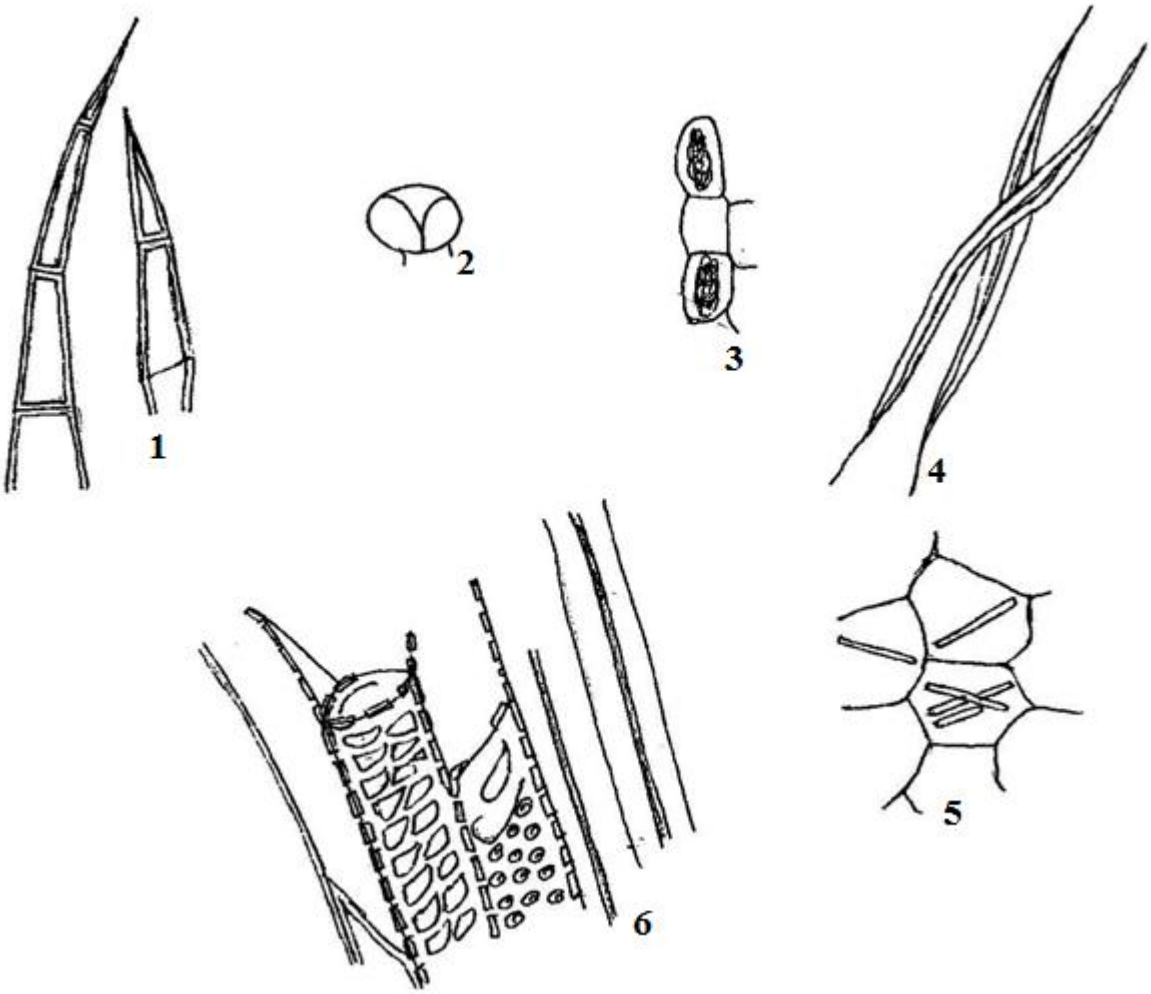


Fig.32. ***Peristrophe bicalyculata*** **powder**: 1. Multicellular uniseriate trichomes, 2.Glandular trichome, 3. Epidermal cells with cystolith, 4. Acicular fibers, 5.Vessels adjoining fibers, 6. Parenchyma with crystals.

3.e.*Polycarpea corymbosa* (L.) Lam.(Caryophyllaceae)

Synonyms: *Achyranthes corymbosa* B.Heyne ex Wall.

Sanskrit: Bhisatta, Okharadi, Parpata, Tadagamritikodbhava.

Vernacular Names:

Hindi : Bugyale, Zutniokhad.

Kannada : Paade Mullu Gida, Poude Mullu, Poude Mullu Gida.

Malayalam : Katu-Mailosina.

Marathi : Koyap, Maitosin.

Tamil : Nilaisedachi, Cataicciver, Pallippuntu.

Telugu : Bommasari, Rajuma.

Distribution and habitat

An annual herb, generally erect and often very strict, often branched from the base but sometimes with simple main stems, 5–38 cm. tall, internodes covered with more or less curled white hairs when young, often glabrescent when older. Leaves opposite or apparently whorled, narrowly linear, acute and then terminating in a hair-like bristle 1 mm. long and caducous, 5–30 mm. long, 0.5–1 mm. broad, 1-nerved, glabrous or nearly so when mature. Inflorescences terminal to branches, of many-flowered cymes, differing greatly in density (see below). Flowers silvery white to pink or purplish red. Sepals lanceolate, acuminate, 2.5–3.75 mm. long, glabrous. Petals about 1.25 mm. long, slightly emarginate or erose. Stamens usually 5, 0.75 mm. long. Ovary with 5–13 ovules; style 0.25 mm. long or even less.

Morphological features

An annual herb, generally erect and often very strict, often branched from the base but sometimes with simple main stems, 5–38 cm. tall, internodes covered with more or less curled white hairs when young, often glabrescent when older. Leaves opposite or apparently whorled, narrowly linear, acute and then terminating in a hair-like bristle 1 mm. long and caducous, 5–30 mm. long, 0.5–1 mm. broad, 1-nerved, glabrous or nearly so when mature. Inflorescences terminal to branches, of many-flowered cymes, differing greatly in density (see below). Flowers silvery white to pink or purplish red. Sepals lanceolate, acuminate, 2.5–3.75 mm. long, glabrous. Petals about 1.25 mm. long, slightly emarginate or erose. Stamens usually 5, 0.75 mm. long. Ovary with 5–13 ovules; style 0.25 mm. long or even less.

Medicinal uses

The plant used for the treatment of inflammation, jaundice, urinary disorders and other kidney problems. The plant also showed the antioxidant activity(Singh *et.al*., 2009). .

Previous Phytochemical reports

The whole plant revealed the presence of alkaloid, catechin, coumarin, flavonoid, tannin, saponin, steroid,phenol, glycoside, terpenoid and xanthoprotein(Nishanthini and Mohan , 2013). Sterols like α-1 barrigenol, camelliagenin A and stigmasterol have been isolated from this plant (Ghazanfar, 1994).

Previous pharmacognostic reports

Very little data (only the T.S. of various parts) available of this plant (Jyothi *et.al*., 2010) but there T.L.S and R.L.S has not been studied. So the plant has been subjected for a detailed study.

Materials and methods

The plant material has been collected from Halol, Gujarat. Phytochemical analysis of the plant for their secondary metabolites such as alkaloids, flavonoids, phenolics, steroids, quinones, mucilages and other compounds were done by standard methods of chromatography and spectrophotometry described in Chapter 2. Pharmacognostic, HPTLC finger printing and physico-chemical studies were done by using standard methods described in Chapter 2.

Results

Phytochemistry

The plant is found to contain flavonoids such as apigenin, acacetin, 3'- OMe luteolin and 7,3-di OMe quercetin. The phenolic acids were vanillic, syringic, ferulic (*cis*- and *trans*-isomers). Mucilage amounted to 7.1% consisting of glucose and xylose. The plant also showed the presence of unidentified alkaloids and steroids.

Pharmacognosy

Root : T.S (Fig.33)

The T.S of the root was circular in outline with a large central woody region. The cork was poorly developed and consisted of 2 to 3 rows of rectangular thin walled cells. The secondary cortex was made up of 5 to 8 rows of comparatively large polygonal compactly packed, thin walled parenchymatous cells wherein the

isolated vascular bundles laid. The vascular bundles were circular and separated by two to three rows of thin walled radialy elongated parenchyma, some of them with reddish brown tanniniferous contents. The vascular bundles were made of outer 2 to 4 layers of phloem followed by 8 to 9 layers of xylem and devoid of medullary rays. The central portion of root were dominated by wood. The phloem were 4 to 6 layers thick and made up of usual elements of phloem. Wood consisted of vessels, tracheids, fibers and rays. Mostly vessels were found solitary and dominated in the centre. Medullary rays were thin walled and radially elongated. Vessels were many distributed throughout or occurring singly or in groups. Vessels and tracheids were simple and boarded pitted. Protoxylem showed annular and spiral thickening.

Root : T.L.S (Fig.34)

Cork cells appeared thin walled rectangular. Parenchyma showed the deposition of reddish brown tanniniferous contents. Fibers were thin walled. xylem rays were thin walled and pitted. The vessels had thick bordered pits with slit like openings.

Root : R.L.S (Fig.35)

Cork cells appeared thin walled rectangular. The cortical parenchyma were polygonal in shape. Ray cells appeared rectangular. The xylem rays were thin walled. Protoxylem showed annular thickening.

Leaf micromorphology

The stomata were of anomocytic type. The stomatal index was 7-10.

Leaf : T.S (Fig.36)

The T.S. of **midrib** showed the large polygonal shaped epidermal cells covered by thick cuticle. The outer walls were thick and papillose. The stomata occurred on both the surfaces and the guard-cells were accompanied by subsidiary cells. The guard-cells were elevated and the front cavity was placed in a depression formed by the papillose outer epidermal walls. The cells of the ground tissues on the both sides were elongated and the lateral walls were wavy, the cells were compactly arranged and some of them were filled with chlorophyll, reddish brown contents and rosette crystals. The central vascular bundle were capped by thick walled sclerenchymatous sheets on both sides. The cells outside the sclerenchymatous sheets contained chlorophyll concentrated towards inner sides. The trichomes were found present on both lower and upper epidermis in traces.

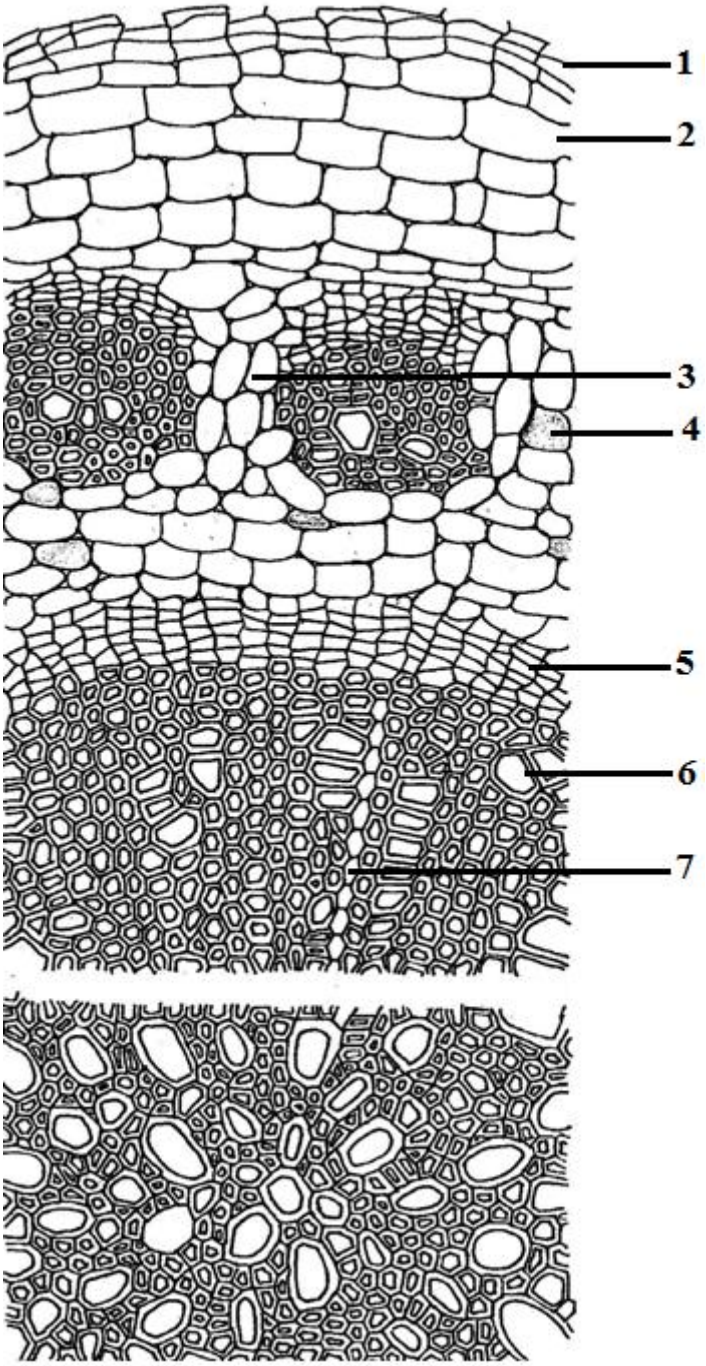


Fig.33. ***Polycarpea corymbosa*** **root, T.S**: 1. Cork, 2. Cortex, 3. Thin walled parenchyma separating two vascular bundles, 4. Parenchyma with reddish brown contents, 5. Phloem, 6. Vessels 7. Xylem rays.

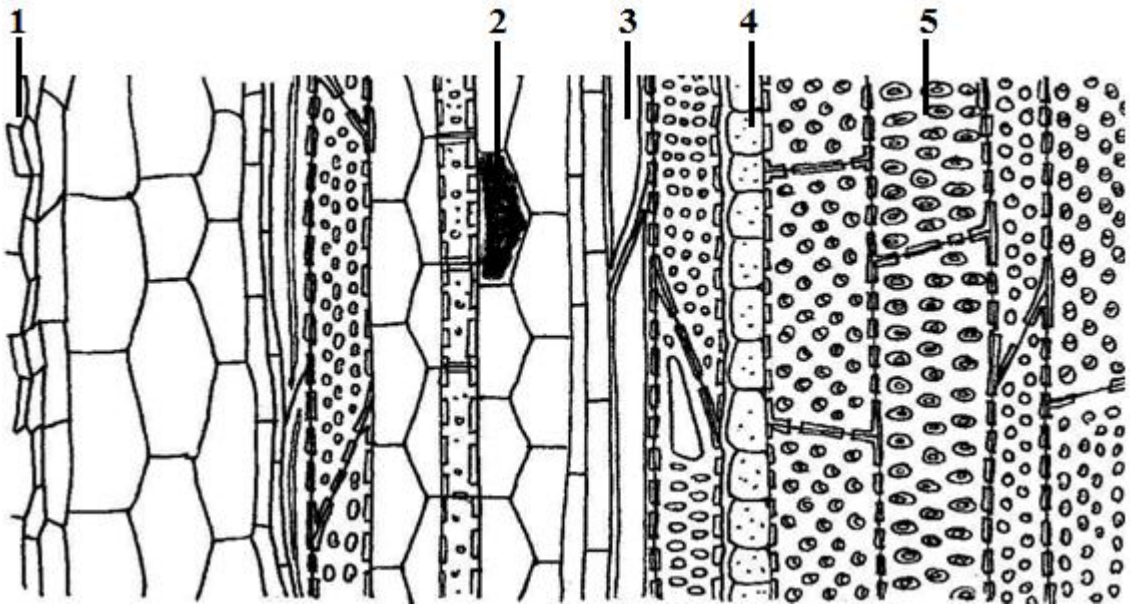


Fig.34. ***Polycarpea corymbosa*****root, T.L.S**:1. Cork, 2. Parenchyma with reddish brown contents, 3. Thin walled fiber, 4. Thin walled xylem rays, 5. Vessels with thick bordered pits.

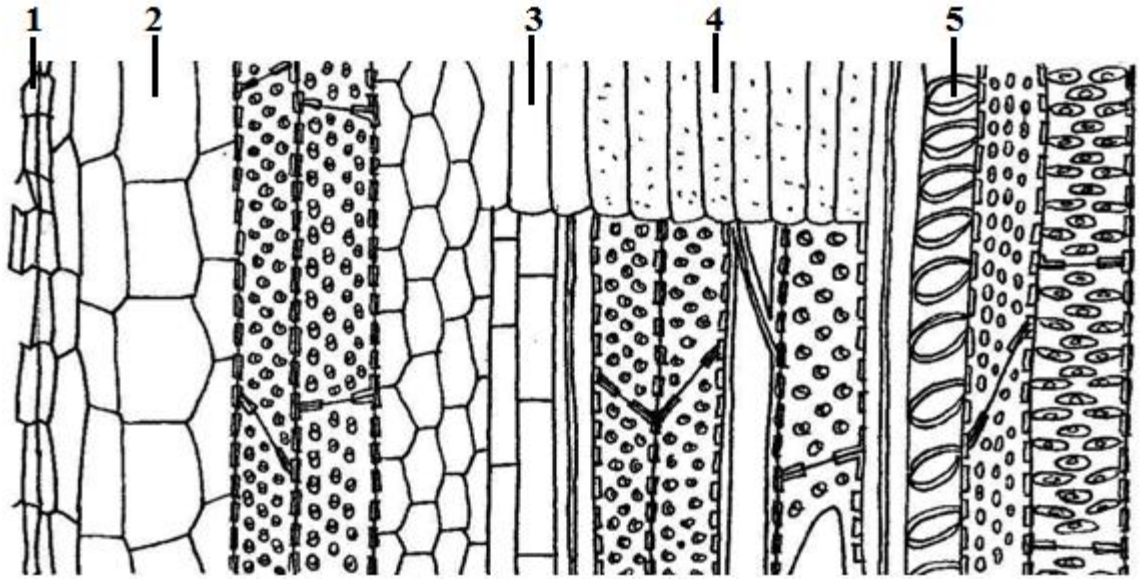


Fig.35. ***Polycarpea corymbosa*****root, R.L.S**:1. Cork, 2. Cortical parenchyma , 3.Phloem rays,4. Xylem rays, 5. Vessels with annular thickenings.

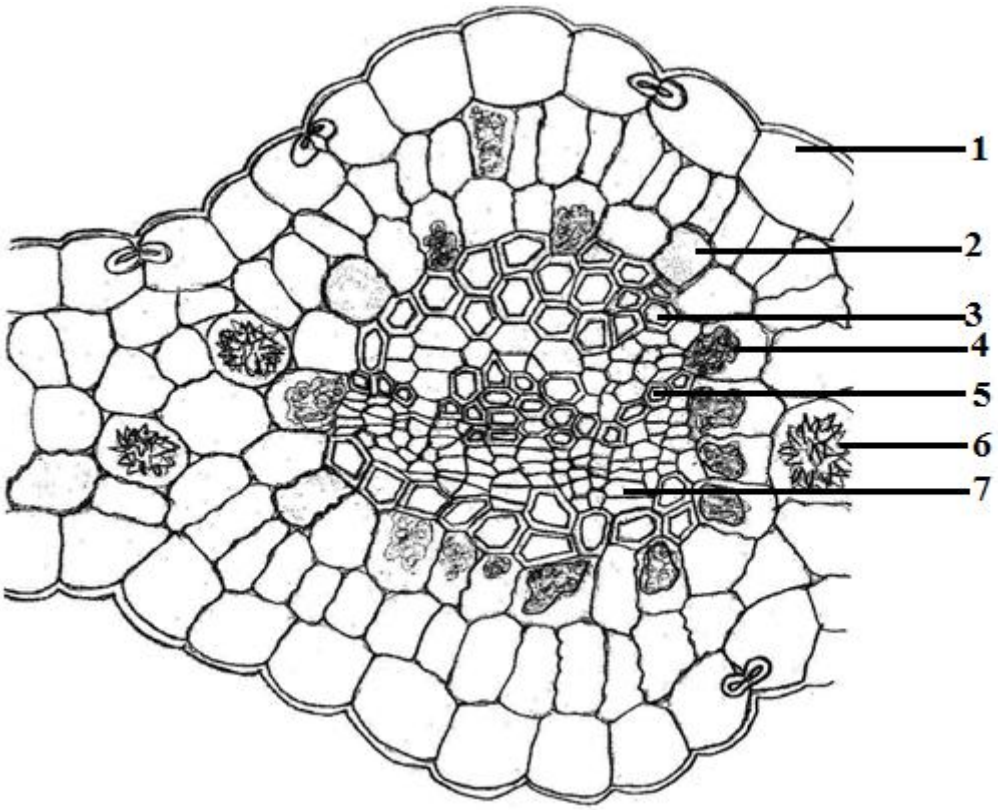


Fig.36. ***Polycarpea corymbosa*** **leaf, T.S:** 1. Epidermis with thick cuticle, 2.Chlorenchyma, 3. Parenchyma with reddish brown contents, 4. Sclerenchyma, 5.Xylem, 6. Rosette crystal, 7. Phloem .

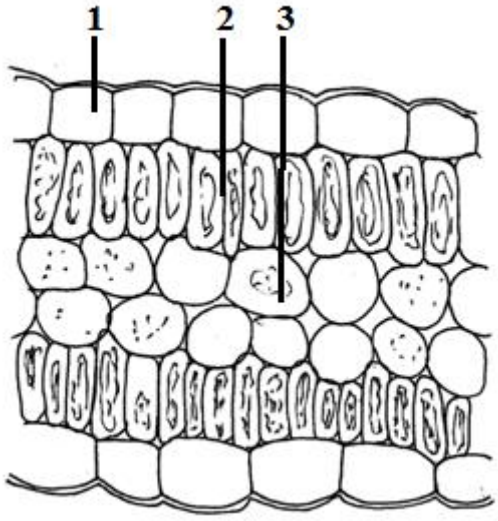


Fig.36.a. ***Polycarpea corymbosa*** **leaf lamina,T.S**: 1. Epidermis with thick cuticle, 2. Palisade cells, 3. Spongy parenchyma.

In the **lamina portion (Fig.36.a)**the epidermal cells were barrel shaped and with thick cuticle. Mesophyll was isobilateral consisted of palisade and spongy tissues. The palisade was single layered and was packed with chloroplasts. The spongy tissues contained loosely arranged parenchyma with intercellular spaces.

Stem : T.S (Fig.37)

The epidermis consisted of polygonal cells covered with a thick cuticle. The outer and inner walls of the cells were greatly thickened. The hypodermis consisted of one or two layers chlorenchyma. The cortex was 3 to 6 layered made up of thin walled tabular cells, many with deposition of reddish brown tanniniferous contents. The pericycle was a continuous ring made up of small rectangular thick walled sclerenchyma with few thick walled parenchyma. The phloem was a narrow zone consisting of usual elements of phloem. Secondary xylem consisted of vessels, tracheids, fibres and wood parenchym. Fibre tracheids were found in patches. The xylem rays were absent. Vessels were boarded pitted. Reticulate thickened vessels and tracheids were also common here. The centre pith was made up of thick walled parenchyma arranged loosely.

Stem : T.L.S (Fig.38)

Epidermal cells were barrel shaped with thick cuticle. The cells of chlorenchyma were upright polygonal adjoining a cells containing reddish brown contents. Fibers were straight and narrow lumened. The phloem cells were also showed the deposition of reddish brown contents. Vessels were boarded pitted and pits arranged alternately followed by reticulate thickened vessels.

Stem : R.L.S (Fig.39)

Epidermal cells showed outer and inner thickened walls. Scleranchyma were thick walled. The fibre tracheids were with narrow lumen and simple pitted. The protoxylem showed spiral thickened vessels. The pith cells appeared large rectangular and were thick walled.

Powder study (Fig.40)

The components present in the powder were parenchyma containing reddish brown contents, branched trichomes, rosette crystals, thick walled narrow lumen fibers and boarded pitted vessel. The branched trichomes found in powder are derived from the flowers present in the material. Leaves contained very rare branched trichomes.

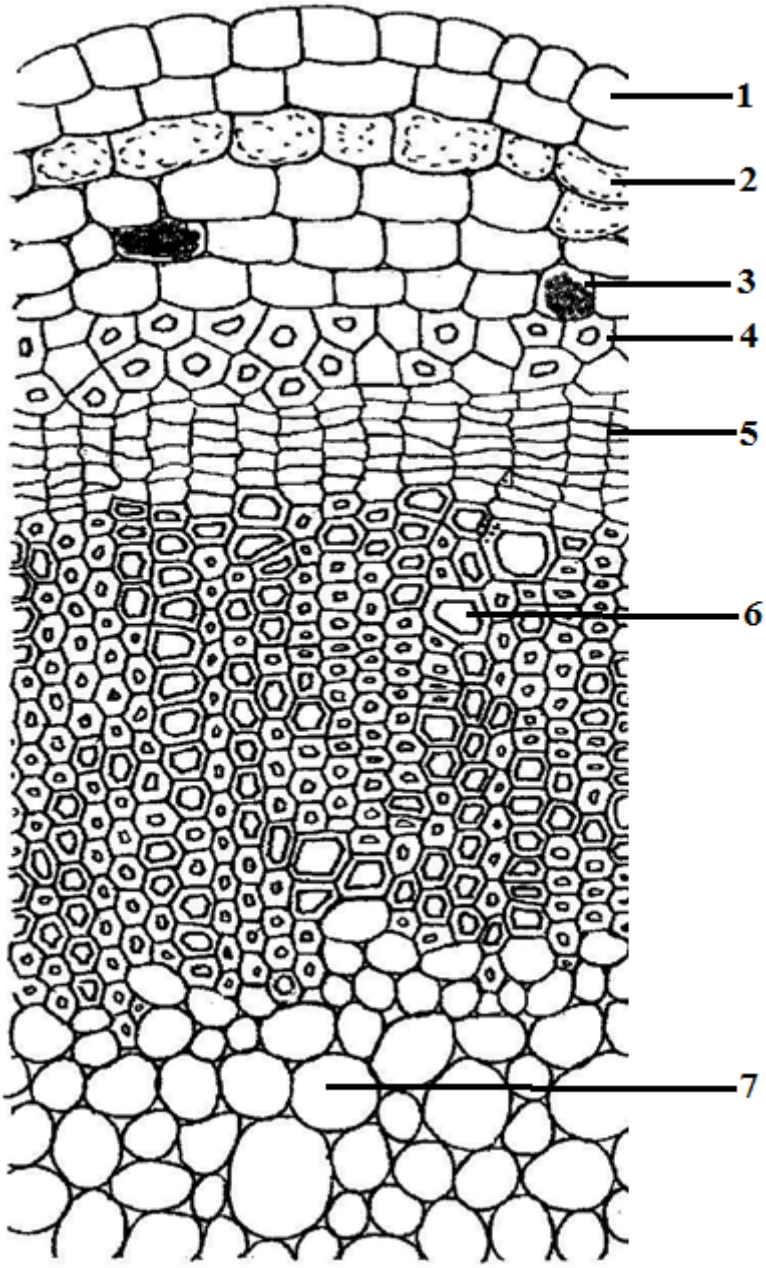


Fig.37. ***Polycarpea corymbosa*****stem, T.S**: 1. Epidermis with thick cuticle, 2.Chlorenchyma, 3. Parenchyma with reddish brown contents, 4. Sclerenchyma, 5.Phloem, 6. vessel, 7. Pith.

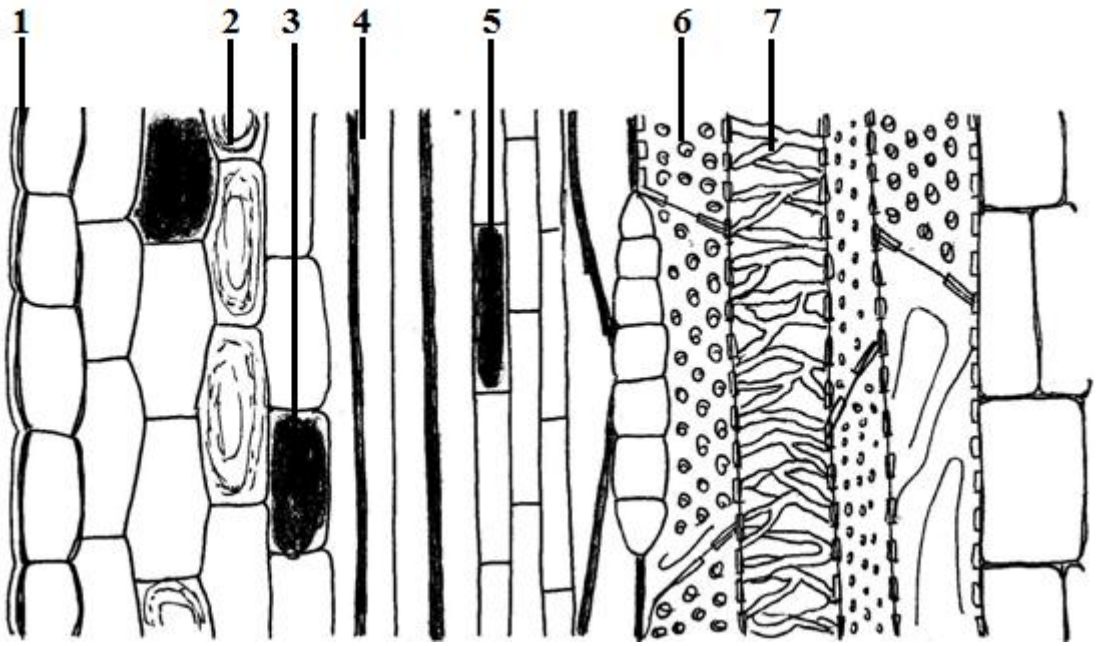


Fig.38. ***Polycarpea corymbosa*** **stem, T.L.S**: 1. Epidermis with thick cuticle, 2.Chlorenchyma, 3. Parenchyma with reddish brown contents, 4. Fiber , 5. phloem with deposition of reddish brown contents, 6. Boarded pitted vessel, 7. Reticulate thickened vessel, 8.Pith.

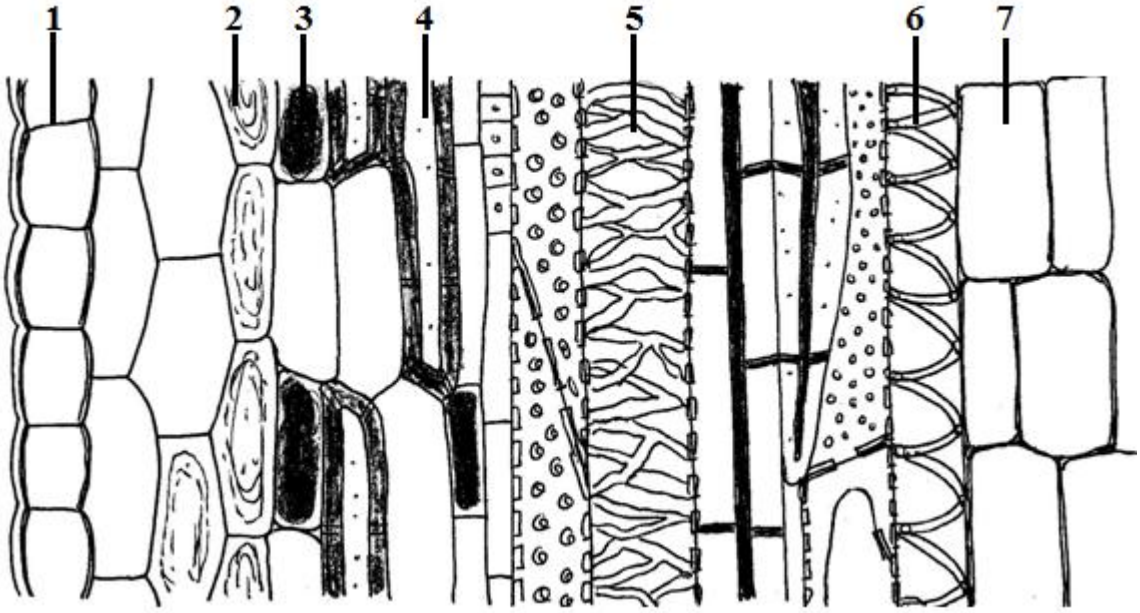


Fig.39. ***Polycarpea corymbosa*** **stem, R.L.S**: 1.Epidermis with thick cuticle, 2.Chlorenchyma, 3. Parenchyma with reddish brown contents, 4. Sclerenchyma, 5.Reticulate thickened vessel, 6. Spiral vessel, 7. Pith.

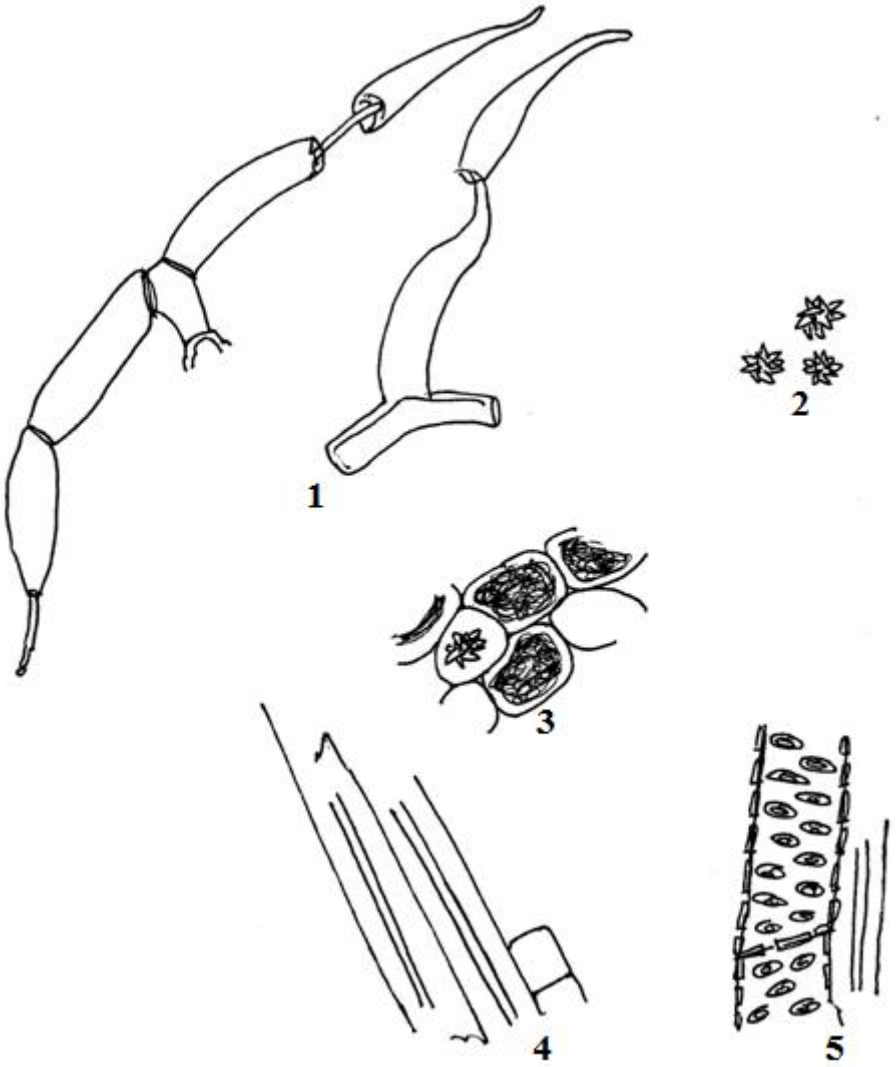


Fig.40. *Polycarpea corymbosa* powder study: 1. Branched trichomes, 2. Rosette crystals, 3. Parenchyma containing reddish brown contents, 4. Thick walled narrow lumened fibers 5.Borded pitted vessel.

Distinguishing features

Pharmacognostic markers :

Root

1. Parenchyma containing reddish brown contents.
2. Absence of rays.
3. Annular and spiral thickened vessels.

Leaf

1. Outer walls of epidermis were thick and papillose.
2. Anomocytic type of stomata.
3. Parenchyma containing reddish brown contents. .
4. Vascular bundle were capped by thick walled sclerenchymatous sheets.
5. Rosette crystals.

Stem

1. The outer and inner walls of the epidermal cells were greatly thickened.
2. The hypodermis was chlorenchymatous.
3. Parenchyma containing reddish brown contents.
4. Pericycle continuous and was sclerenchymatous.
5. Absence of rays.
6. Reticulate thickened vessels.
7. Pith parenchyma thick walled.

Phytochemical markers

1. Apigenin.
2. Acacetin.
3. 3'- OMe luteolin.
4. 7,3-di OMe quercetin.

Physico-chemical analysis:

Table: 6. Values obtained for the proximate analysis.

Sr.No.	Parameter	Mean ± SD (%)*			Average (%)
		Summer	Monsoon	Winter	
1.	Total Ash Content	4.69±0.27	4.92.86±0.23	4.98±0.19	4.86
2.	Acid Insoluble Ash content	0.49±0.07	0.42±0.07	0.48±0.09	0.46
3.	Alcohol soluble extractives	4.29±0.23	3.92±0.38	4.63±0.09	4.28
4.	Water soluble extractives	15.34±0.14	15.02±0.28	15.09±0.20	15.15

*Each value is a mean of 3 reading

3.f. *Rungia repens* (L.) Nees.(Acanthaceae)

Sanskrit : Parpata, Parpatha.

Vernacular Names:

Gujarati : Parpat.

Hindi : Kharmor, Kharmar.

Kannada : Kodagasaale Gida, Kodaga Saale Gida.

Marathi : Ghatipitpapada.

Tamil : Kodagasalai, Kotacuri, Cataikkaranti, Kotaculi, Kotakacalai, Maram.

Telugu : Palakavelli.

Distribution and habitat

The plant is a spreading decumbent herb found throughout India mostly as a weed in moist places. (Gamble 1921, Saxena *et.al*, 1995).

Morphological features.

Stems usually decumbent, often rooting near the base, then erect, slender, subterete, glabrous or puberulous. Leaves upto 5 cm long, subsessile or shortly petioloate, oblong-lanceolate, acute, lineolate on both sides, glabrous or nearly so, base usually tapering, less commonly rounded and unequal-sided; main nerves about 6 pairs; petioles rarely reaching upto 0.4 cm long. Flowers in erect terminal usually pubescent, imperfectly 1-sided spikes, upto 8 cm long; bracts broadly elliptic, pubescent, cuspidate, much imbricate, the margins thinly scarious, ciliate; bracteoles linear-lanceolate, acute, with scarious margins, minutely pubescent. Calyx puberulous, divided to the base; segments lanceolate-subulate. Corolla 2-lipped, 1.5 cm long; upper lip oblong, emarginated; lower lip shortly 3-lobed. Stamens 2; anthers 2-celled, the cells often superpose, the lower cell often with a white basal appendage, lower anther cells with a white appendage at the base. Disk annular or shortly copular. Ovary 2-celled; ovules 2 in each cell; style filiform; stigma minutely 2-fid. Capsules ovoid-oblong, acute, compressed, with scarious faces and hard edges, pubescent. Seeds suborbicular, rugose with concentric furrows, pale-brown.

Action and uses:

The herb is used in the treatment of cough and fever and is also credited with vermifugal and diuretic properties (Trease and Evans, 2002). Fresh, bruised leaves are mixed with castor oil and applied to scalp to cure *Tinea capitis* , a scaly fungoid

infection, usually occurring amongst children (Anon.1996, Anon.1999, Kirtikar and Basu, 1994, Nadkarni and Nadkarni,2002).The juice of the leaves is considered cooling and aperients, and is given to children suffering from smallpox. Bruised leaves are applied to relieve pain and reduce swelling. In Bihar, the roots are used as a febrifuge by the tribal population (Anon.1996,Trease and Evans, 2002). There are also reports that it is of use as diuretic and vermifuge (Anon.1999).

Previous Phytochemical reports

The hydroalcoholic extract of leaf is found to contain phytosterols, terpenes, tannins, flavonoids and carbohydrates(Swain *et al.*,2008).Investigation on the flavonoid pigments in ivory-white and pale yellow flowers showed the presence of luteolin and chrysoeriol (3'-O-methylluteolin) and their glucosides (Sankar *et.al*,1964) . Flowers with deep yellow tubular portion and bluish pink spots contain isosalipurposide (2'-glucosyloxy-4,4',6'-trihydroxychalcone), luteolin and its 7-glucoside. The bluish pink colour is due to the presence of delphinidin-3,5-diglucoside(Seshadri and Vydeeswaran,1972).

Previous pharmacognostic reports:

Very little data available on pharmacognosy of this plant. Only T.S of various parts of the plant has been studied (Jyothi *et.al.*, 2010).

In the present work roots, stem and leaves of this plant has been subjected to phytochemical and pharmacognostic studies.

Materials and methods

The plant material was collected from Timbi in outskirts of Baroda, Gujarat. Phytochemical analysis of the plant for their secondary metabolites such as alkaloids, flavonoids, phenolics, steroids, quinones, mucilages and other compounds were done by standard methods of chromatography and spectrophotometry described in Chapter 2. Pharmacognostic, HPTLC finger printing and physico-chemical studies were done by using standard methods described in Chapter 2.

Results

Phytochemistry :

The plant is found to contain flavonoids such as quercetin, 7'- methoxy quercetin and kaempferol. The phenolic acids were vanillic, syringic, ferulic (*cis*- and *trans*-isomers), gentisic and protocatechuic acids. The flowers of the plant contained the anthocyanin delphinidin , flavonol kaempferol, and its 4'-OMe

derivative, while the phenolic acids located were vanillic, *p*-coumaric and *p*-hydroxybenzoic acids. Mucilage amounted to 7.3% consisting of ribose and xylose. The plant also showed the presence of unidentified alkaloids and steroids.

Pharmacognosy

Root : T.S (Fig.41)

The T.S of the root was circular in outline with a large central woody region. The cork was poorly developed and consisted of 2 to 4 rows of rectangular to slightly tangentially elongated thick walled cells. The secondary cortex was very narrow consisting of two zones, outer zone of two to four rows of comparatively large polygonal compactly packed, thin walled parenchymatous cells and inner small rectangular cells; many of which were found filled with rosette crystals(6-8μm). Endodermis was prominent and thick walled. Some of the phloem parenchyma cells at the outer region contained rosette crystals. Wood consisted of vessels, tracheids, fibers and rays. Outer wood was dominated by vessels and tracheids while inner by wood fibers. Medullary rays were thin walled and radially elongated. Vessels were many distributed throughout or occurring singly or in groups.

Root : T.L.S (Fig.42)

Cork cells appeared rectangular with wavy walls. The medullary rays were compressed spindle shaped and thin walled and simple pitted. Fibers showed scanty pits on their walls. The vessels had 3-4 rows of alternate bordered pits with slit like openings.

Root : R.L.S (Fig.43)

The phloem parenchyma contained rosette crystals. Ray cells appeared rectangular. The xylem rays were pitted. Here the vessels were more in number and in a groups surrounded by fiber tracheids from both the sides.

Leaf micromorphology

The stomata were of diacytic type. The stomatal index was 16-18. The trichome index was 9-12. The trichomes were thick-walled unicellular as well as multicellular uniseriate showing broad basal cell, blunt tip and the warty walls. Unicellular and glandular trichomes were rare.

Leaf : T.S (Fig.44)

The **midrib portion** was characterized by a concave bulge on the upper side and hemispherical bulge on the lower side. The epidermal cells were polygonal in

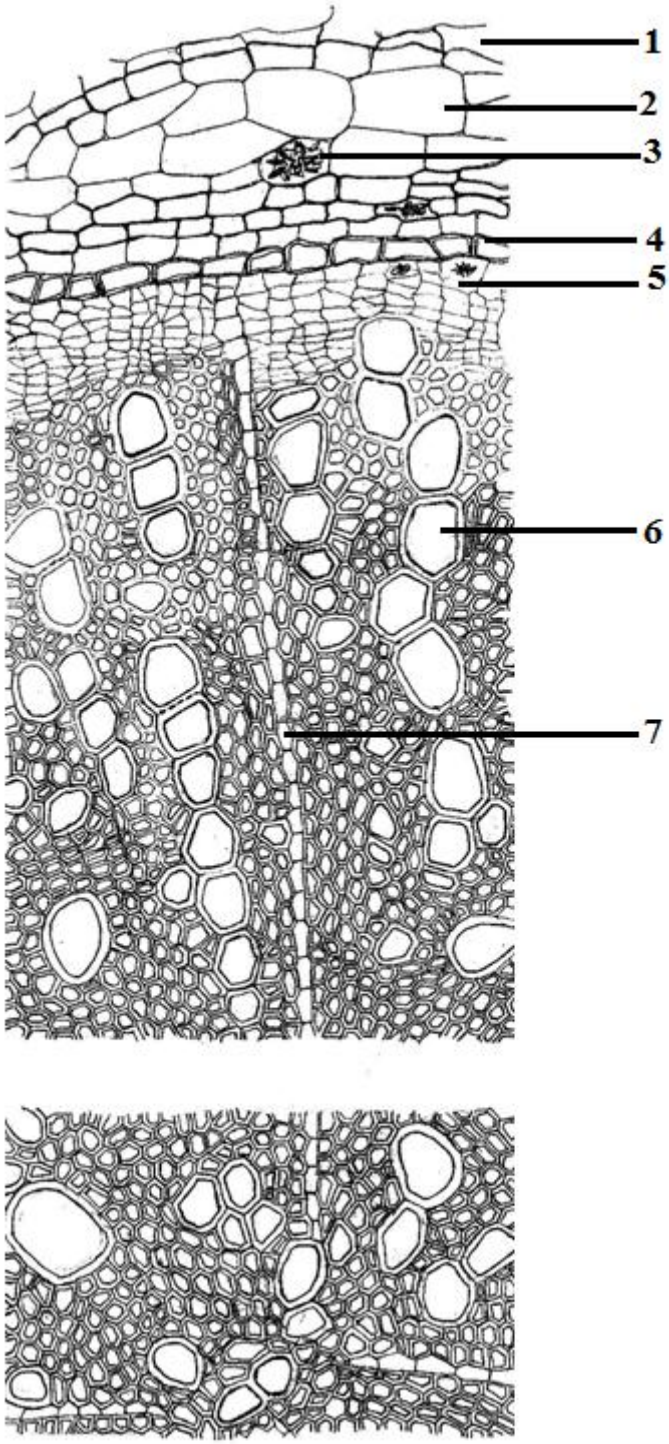


Fig.41. ***Rungia repens*** **root, T.S :** 1. Cork, 2. large polygonal Parenchyma, 3. Parenchyma with rosette crystals, 4. Endodermis, 5. Phloem with rosette crystals, 6. Vessels 7. Xylem rays.

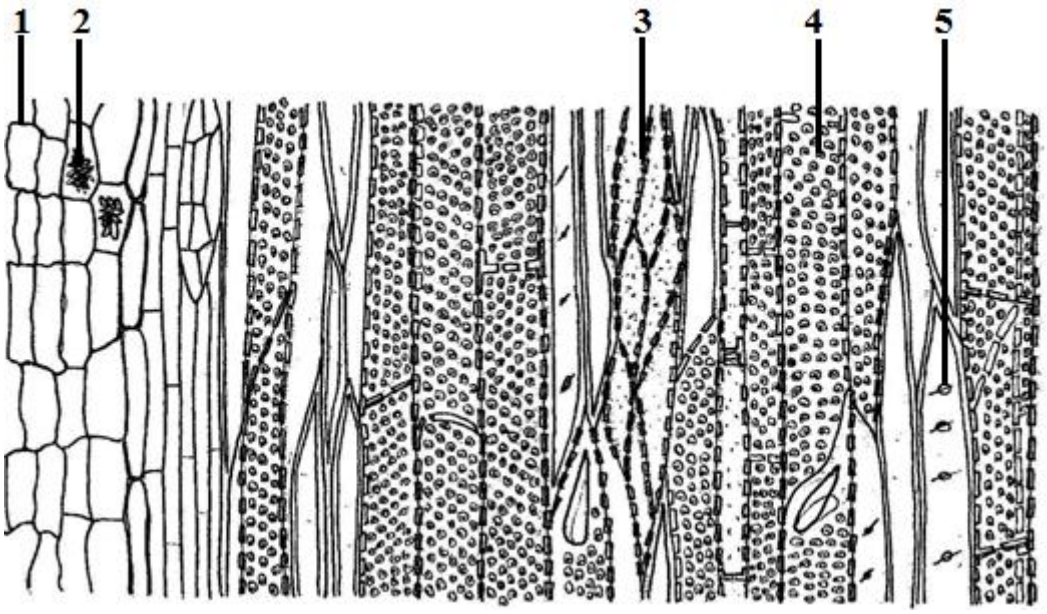


Fig.42.*Rungia repens* root, T.L.S:1. Cork cells with wavy walls, 2. Rosette crystal, 3.Compressed spindle shaped medullary rays,4. Vessels with alternate bordered pits, 5. Fibers with scanty pits.

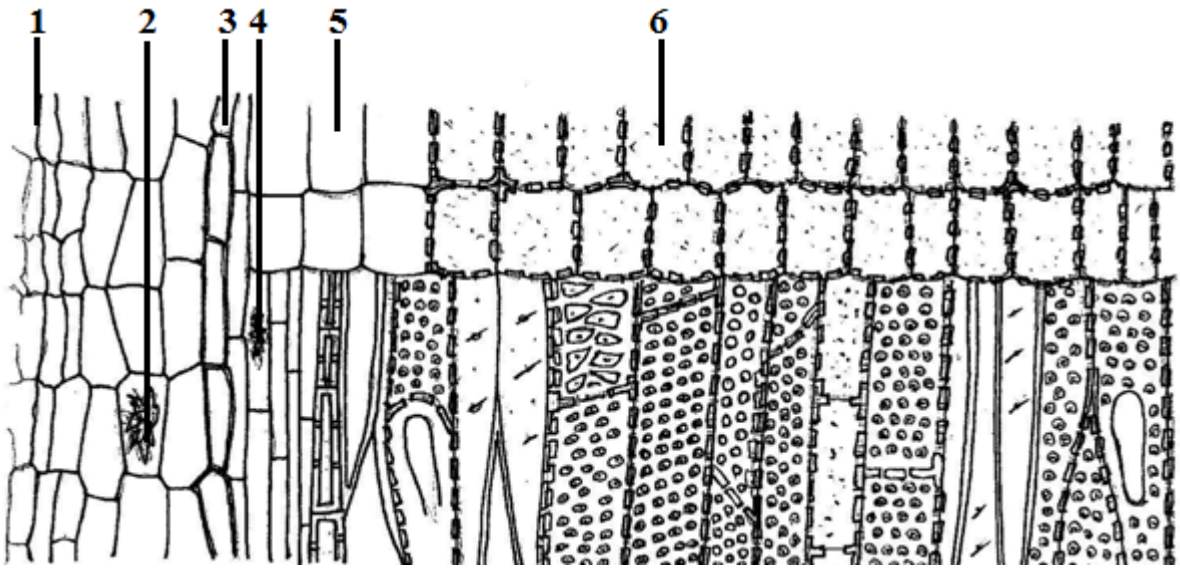


Fig.43. *Rungia repens* root, R.L.S:1. Cork cells, 2. Rosette crystal, 3.Thick walled endodermis, 4. Phloem with rosette crystals, 5. Phloem rays, 6. Xylem rays.

shape covered by thick cuticle. Some of the cells were showed the deposition of globules. The hypodermis on both upper and lower regions made up of angular collenchyma. The ground tissue was parenchymatous and the cells on the upper side were rounded to hexagonal and compactly arranged. The vascular bundle was crescent shaped with 4-7 rows of tracheids, followed by 5-7 layers of phloem. Below this were large parenchymatous cells. The trichomes were found present on both lower and upper epidermis.

In the **lamina portion** the epidermal cells were barrel shaped and some of the cells showed deposition of globules. The mesophyll consisted of palisade and spongy tissues. The palisade was single layered and was finely packed with chloroplasts. The spongy tissues contained loosely arranged parenchyma with intercellular spaces.

Stem : T.S (Fig.45)

The epidermis consisted of polygonal cells covered with a thin cuticle and contained thick-walled unicellular as well as multicellular uniseriate trichomes with broad basal cells. The tip of the trichomes were blunt and the walls were warty. Cystoliths were found present in epidermis at regular intervals. Some of the epidermal cells showed the deposition of globules. The hypodermis was 3-4 layered made up of lamellar collenchyma. The cortex consisted of 5-7 layers of large elongated parenchyma, few of these cells were typical pitted sclerenchymatous and found in a pairs wherein the adjoining walls were straight. The endodermis was single layered and side walls of the cells showed the casparian thickenings. The pericycle was a discontinuous ring made up of small rectangular thick walled cells. The phloem was a narrow zone consisting of usual elements of phloem. Secondary xylem consisted of vessels, tracheids, fibres and xylem rays. Tracheids and fibre layers were intercepted by the continues rows of

vessels. The xylem rays were uniseriate to triseriate. The protoxylem in the Pith region was surrounded by the thin walled pitted parenchyma. The central region of pith contained compactly arranged isodiametric thin walled parenchyma.

Stem : T.L.S (Fig.46)

Epidermal cells were rectangular bearing trichomes and cystolith followed by thick walled collenchyma. The cells of endodermis were polygonal in shape and the side walls of which were thickened. The phloem cells were rectangular in shape. Xylem rays were pitted and biseriate. The fibre tracheids were with broad lumen. The

pith cells appeared squarish to rectangular, thin walled and some of these cells showed pits on their walls. Protoxylem was very prominent with spiral thickening.

Stem : R.L.S (Fig.47)

Cortical sclerenchyma were polygonal, thick walled and with simple pits, lying one above the another in a rack. Xylem rays also were polygonal and thick walled. The walls of vessels were with bordered pits and also showed annular thickening.

Powder study (Fig.48)

The components present in the powder were thick-walled unicellular as well as multicellular uniseriate trichomes having broad basal cell and warty walls with blunt tip, fragmentes of parenchyma with diacytic stomata ,sclereids, thin walled cortical parenchyma, broad lumen fiber, ray parenchyma, fiber tracheids, boarded pitted and spiral vessel.

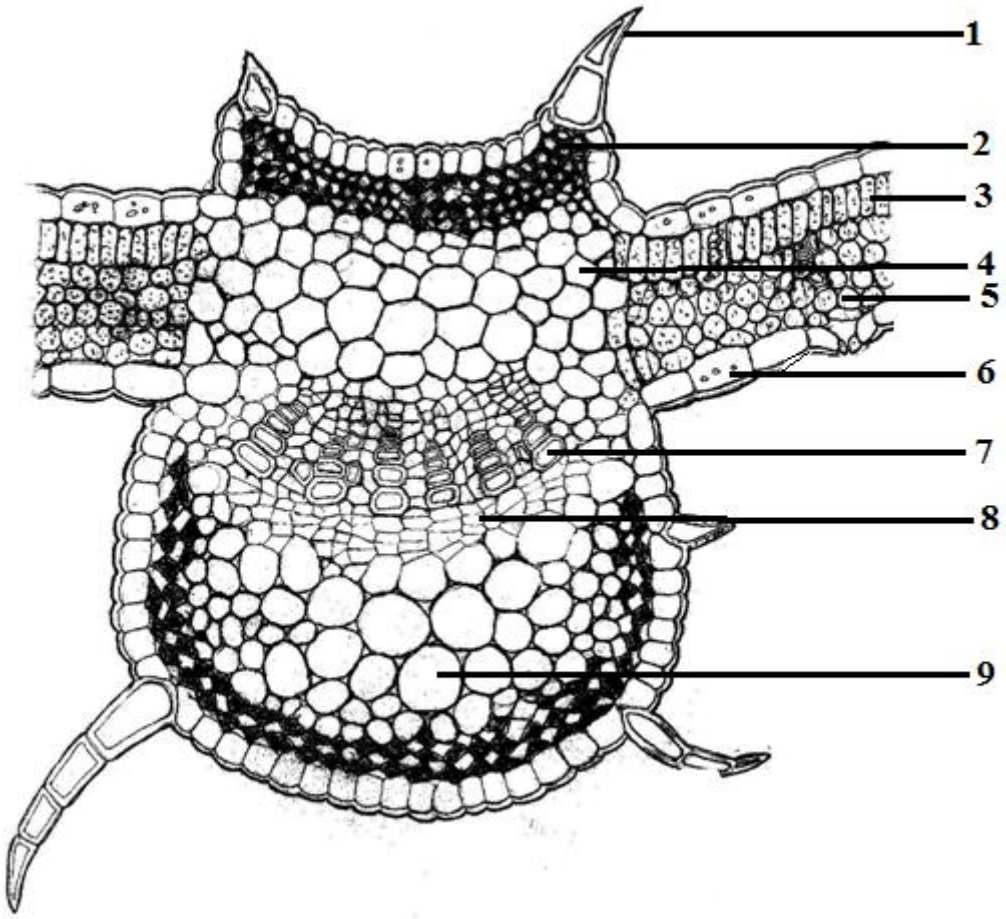


Fig.44.*Rungia repens* leaf, T.S: 1. Trichome with broad basal cell, 2. lamellar collenchyma, 3. Palisade, 4. Hexagonal parenchyma, 5. Spongy parenchyma, 6.Epidermis with deposition of globules, 7. Xylem, 8. Phloem, 9.Ground tissue.

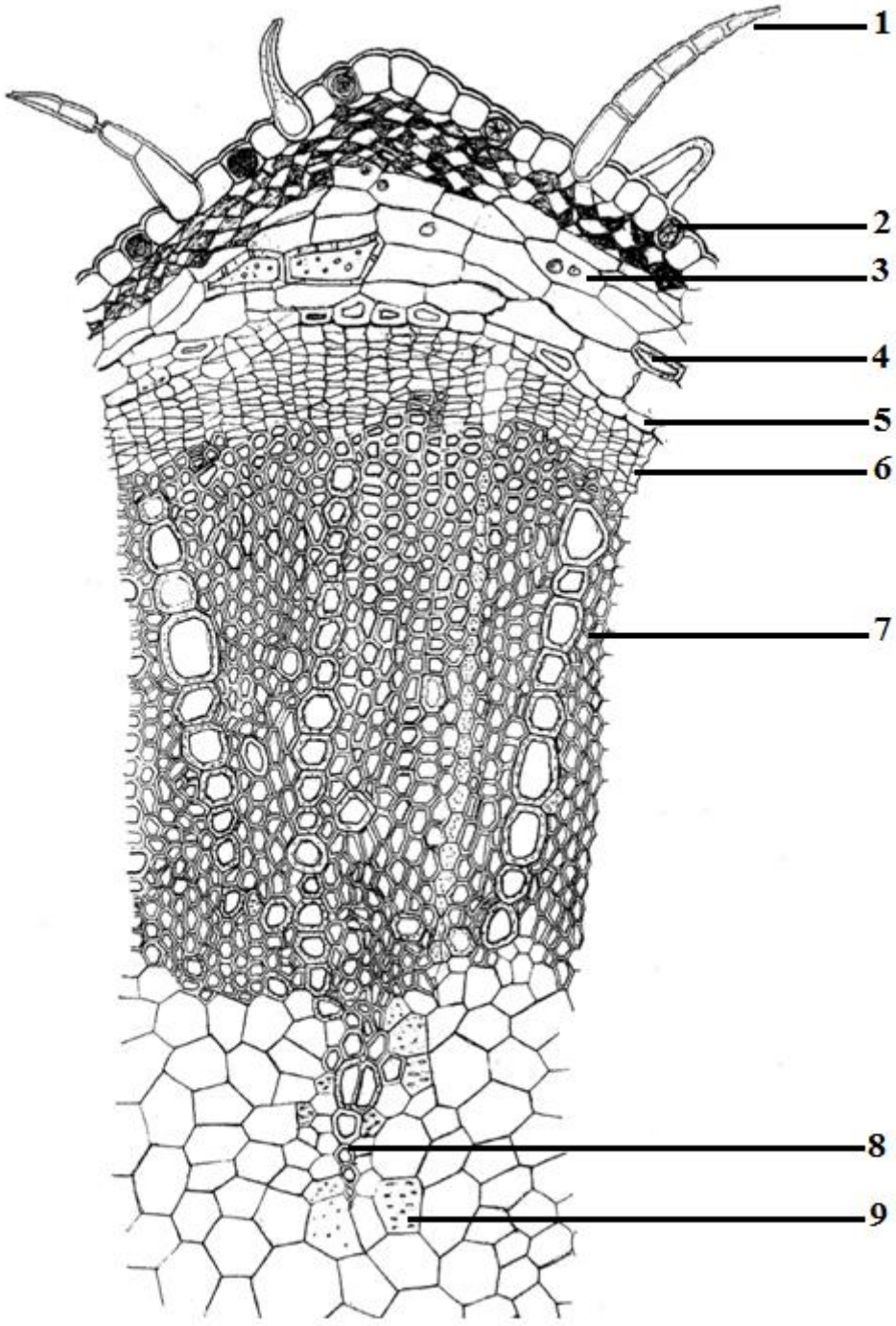


Fig.45.*Rungia repens* stem, T.S: 1. Multicellular uniseriate trichome 2.Epidermal cell with cystolith, 3. Globules in parenchyma cells. 4. Sclerenchyma, 5. Pericycle, 6.Phloem, 7. xylem, 8. Protoxylem, 9.Pitted parenchyma.

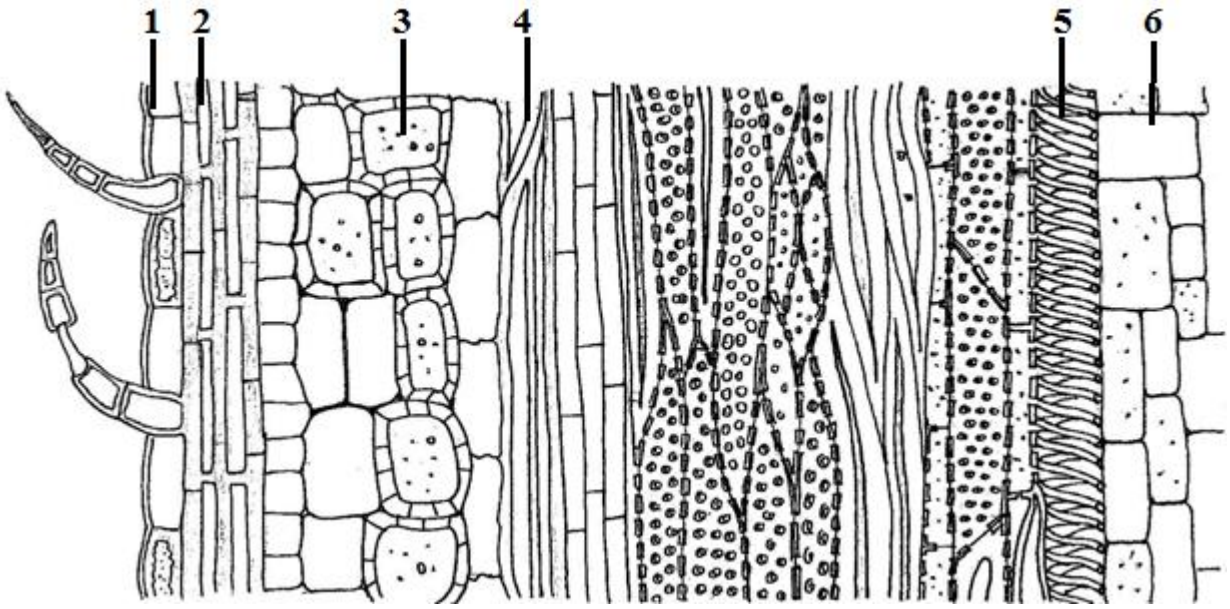


Fig.46.*Rungia repens* stem, T.L.S: 1.Epidermal cell with cystolith and hair, 2.collenchyma. 3. Sclerenchyma, 4. Pericyclic fibers, 5. Protoxylem, 6. Pitted parenchyma.

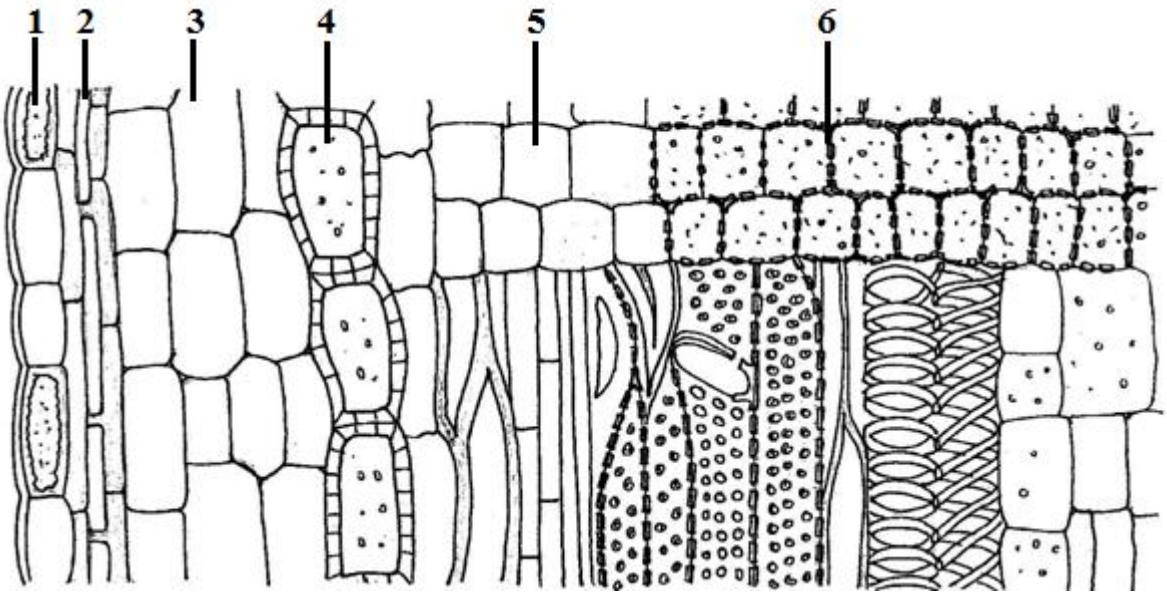


Fig.47. *Rungia repens* stem, R.L.S:1.Epidermal cell with Cystolith, 2.Collenchyma, 3.Parenchyma, 4. Sclerenchyma, 5.Phloem rays, 6.Pitted xylem rays.

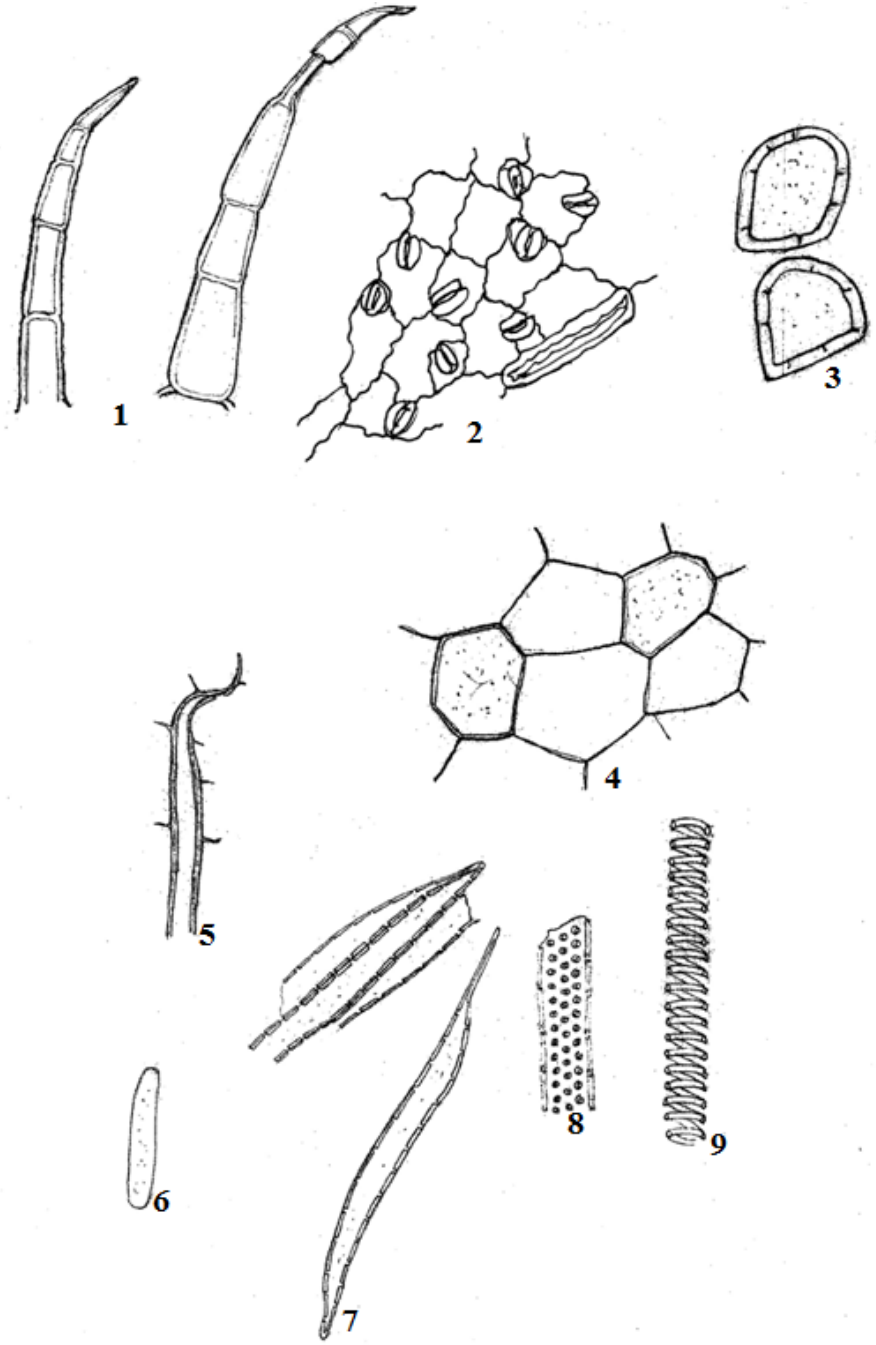


Fig.48.*Rungia repens* , powder study: 1.Trichomes, 2. Epidermal fragment with stomata, 3.Sclereids, 4.Cortical parenchyma, 5. Broad lumened fiber,6. Ray parenchyma, 7.Fiber tracheids, 8. Boarded pitted vessel, 9. Spiral vessel.

Distinguishing features

Pharmacognostic markers :

Root

4. Cortical parenchyma cells bearing rosette crystals.
5. Thick walled prominent endodermis.
6. Phloem parenchyma cells containing rosette crystals.

Leaf

6. Deposition of cystoliths and globules in the epidermis.
7. Diacytic type of stomata.
8. Thick-walled unicellular as well as multicellular uniseriate trichomes with broad basal cell, blunt tip and warty walls.

Stem

8. The epidermal cells showed the deposition of globules.
9. Thick-walled unicellular as well as multicellular uniseriate trichomes with broad basal cell, blunt tip and warty walls.
10. Hypodermis 3-4 layered of lamellar collenchyma.
11. Pitted schlerenchyma in a pairs wherein the adjoining walls were straight

Phytochemical markers

1. Kaempferol.
2. Quercetin.
3. 7'- methoxy quercetin.
4. 4'-OMe kaempferol.
5. Ferulic (*cis*- and *trans*-isomers) acid.

Physico-chemical analysis:

Table:7. Values obtained for the proximate analysis.

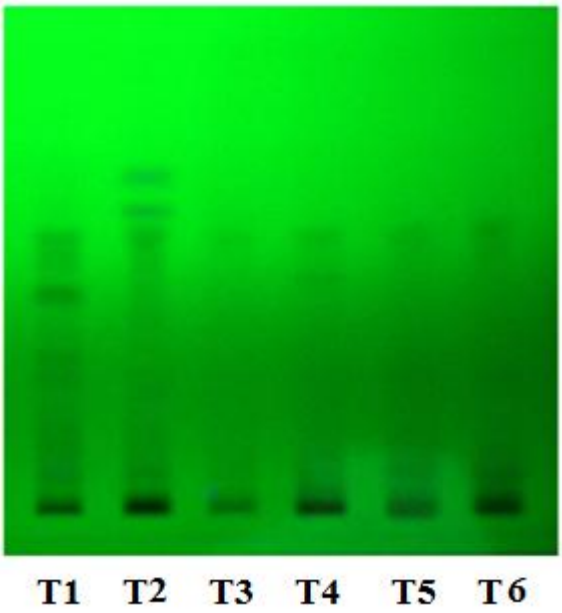
Sr.No.	Parameter	Mean ± SD (%)*			Average (%)
		Summer	Monsoon	Winter	
1.	Total Ash Content	12.28±0.33	12.86±0.06	12.32±0.29	12.49
2.	Acid Insoluble Ash content	0.98±0.05	1.04±0.04	0.92±0.1	0.98
3.	Alcohol soluble extractives	14.47±0.83	13.62±0.91	13.66±0.89	13.92
4.	Water soluble extractives	18.46±0.42	18.33±0.34	18.30±0.30	18.36

*Each value is a mean of 3 reading

3.g. HPTLC fingerprinting and Physo-chemical analysis of *Fumaria parviflora* and its substitutes/adulterants

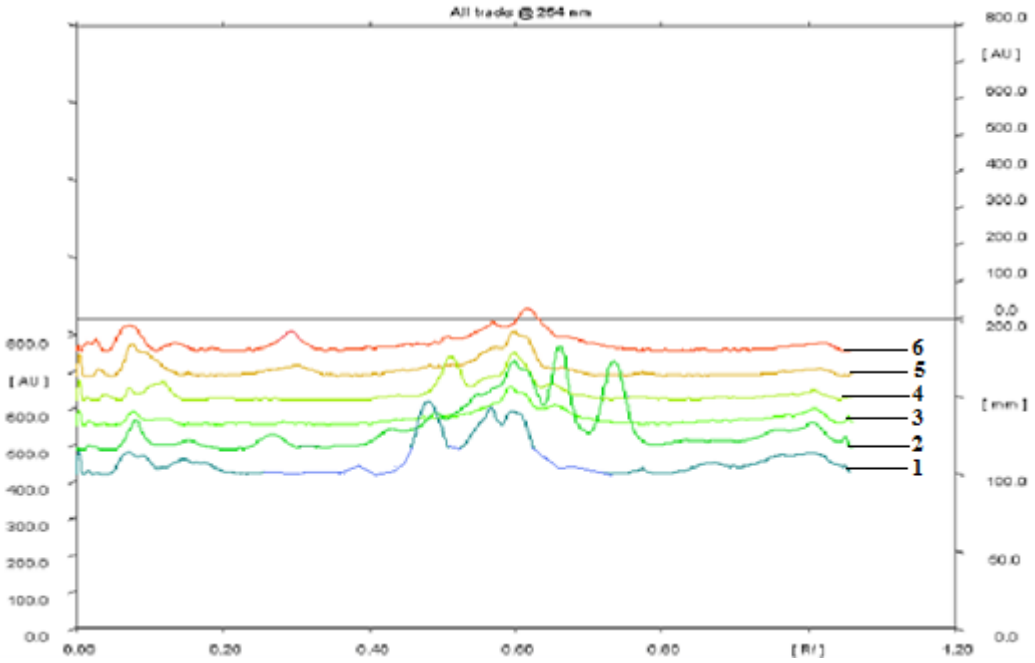
HPTLC fingerprinting

Figure 49.a : HPTLC chromatogram of *Fumaria parviflora* and its substitutes/adulterants. (UV 254 nm).



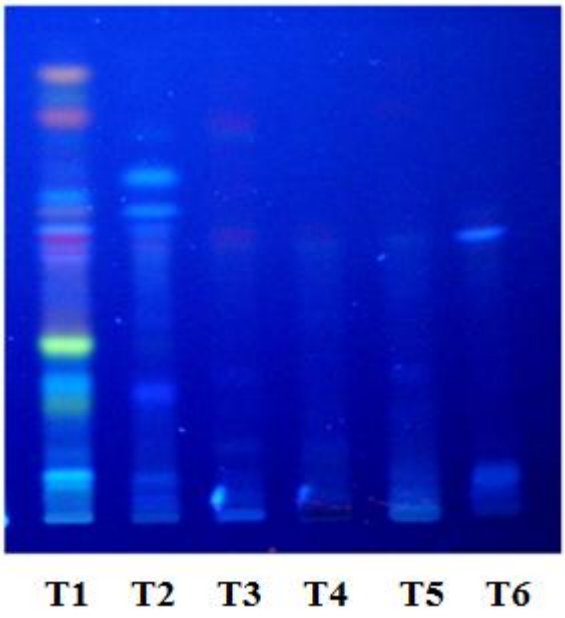
(a).T1-*Fumaria parviflora*, T2- *Justicia procumbens*,T3-*Rungia repens*,T4-*Polycarpea corymbosa*, T5-*Peristrophe bicalyculata* ,T6-*Oldenlandia corymbosa.*

Figure 49.b: HPTLC chromatogram of *Fumaria parviflora* and its substitutes/adulterants.



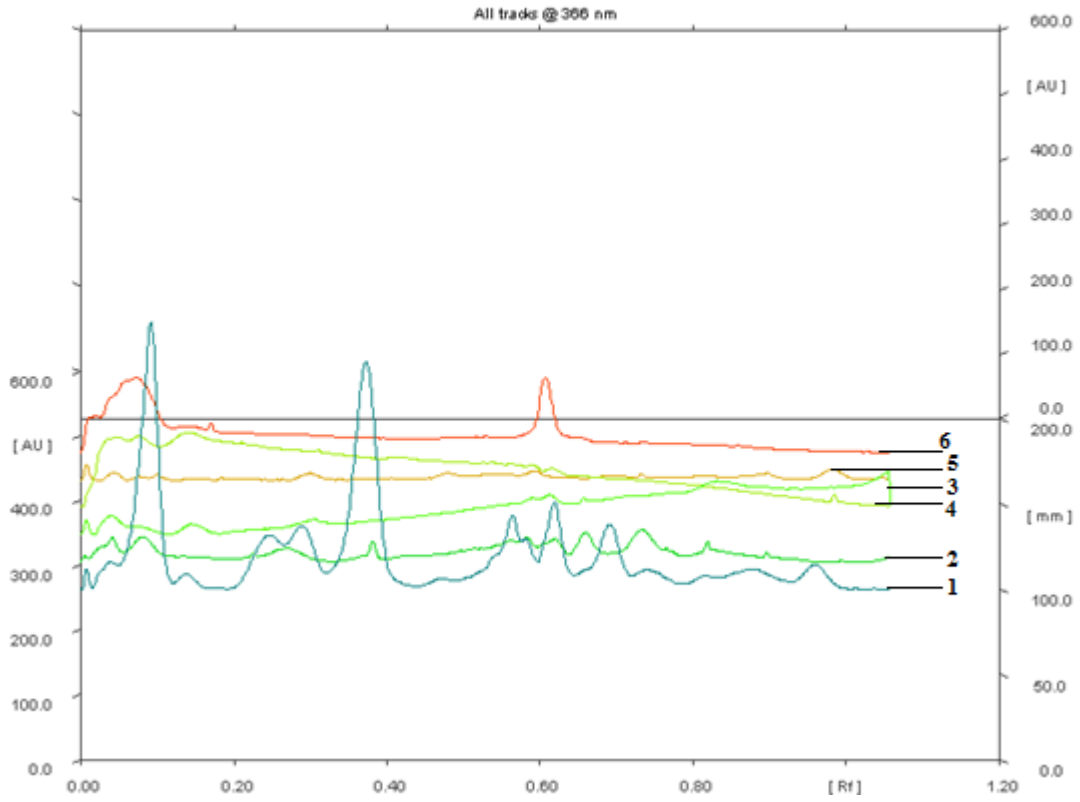
(b).1-*Fumaria parviflora,* 2-*Justicia procumbens,*3-*Rungia repens,*4-*Polycarpea corymbosa,* 5-*Peristrophe bicalyculata* ,6-*Oldenlandia corymbosa.*

Figure 50.a: HPTLC chromatogram of *Fumaria parviflora* and its substitutes/adulterants (UV 366 nm).



(a).T1-*Fumaria parviflora,* T2-*Justicia procumbens,*T3-*Rungia repens,*T4-*Polycarpea corymbosa,* T5-*Peristrophe bicalyculata* ,T6-*Oldenlandia corymbosa.*

Figure 50.b: HPTLC chromatogram of *Fumaria parviflora* and its substitutes/adulterants (UV 366 nm).



(b).1-*Fumaria parviflora*, 2-*Justicia procumbens*,3-*Rungia repens*,4-*Polycarpea corymbosa*, 5-*Peristrophe bicalyculata* ,6-*Oldenlandia corymbosa.*

HPTLC profile of *Fumaria parviflora* showed the presence of 13 peaks when observed under UV 254 nm (fig.49.b)and 15 peaks when observed under UV 366 nm (fig.50.b). There were 3 major peaks found under UV 254 nm at R_f 0.48, R_f 0.56 and R_f 0.59 while 2 under UV 366 nm at R_f 0.09 and R_f 0.37. The *Justicia procumbens* showed the presence of 14 peaks and *Rungia repens* 8 peaks while *Polycarpea corymbosa, Peristrophe bicalyculata* and *Oldenlandia corymbosa* showed the presence of 9 peaks when observed under UV 254 nm(fig.49.b) while under UV366 nm (fig.50.b); *J. procumbens, R. repens, P. corymbosa, P. bicalyculata* and *O. corymbosa* showed the 12,9,7,5 and 3 peaks respectively .

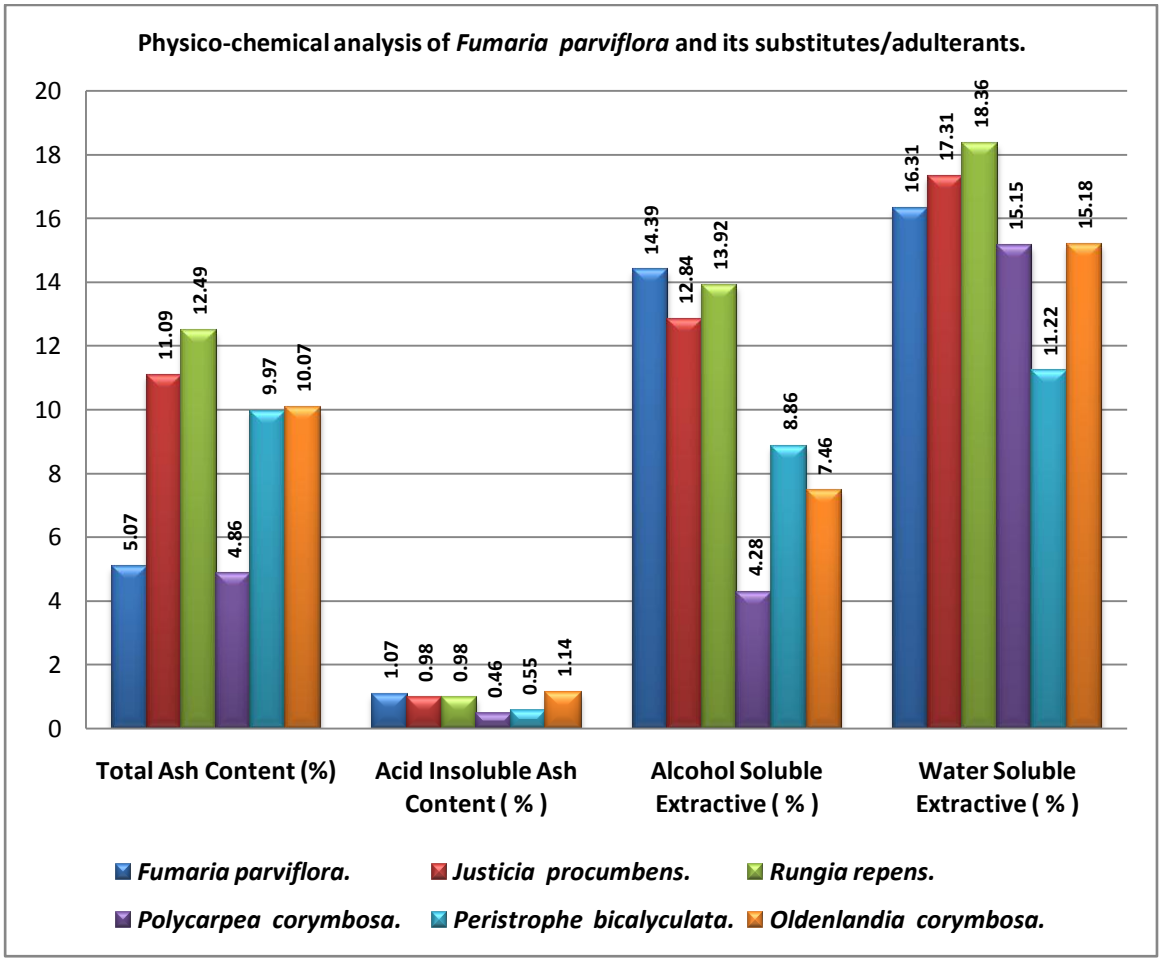
HPTLC profile of *F. parviflora* and its substitute/adult observed under UV 254 nm (fig.49.b) showed that *J. procumbens* was similar in 3 peaks but differed in 11peaks, *R. repens* was similar in 1 peak and differed in 6 peaks. *P. corymbosa*

similar in 1 peaks but differed in 8 peaks while *P. bicalyculata* and *O. corymbosa* were similar in 2 peaks but differed in 7 peaks.

HPTLC profile of *F. parviflora* and its substitute/adultarunts observed under UV 366 nm (fig.50) showed that *J. procumbens* was similar in 4 peaks but differed in 8 peaks, *R.repens* was similar in 3 peaks and differed in 6 peaks. *P. corymbosa* similar in 2 peaks but differed in 5 peaks and *P. bicalyculata* similar in 2 peaks but differed in 3 peaks while *O.corymbosa* was not show any peaks similar to that of *F. parviflora* but differed in having 3 peaks.

Physico-chemical analysis

Physico-chemical analysis of *Fumaria parviflora* and its substitutes/adulterants.



Total ash content

Total Ash Content of *Fumaria parviflora* (5.07 %) along the material collected in different season does not show significant variation (Table-2) while the closest value to the substitute/adulterant is *Polycarpea corymbosa* (4.86 %). Other substitute/adulterant have higher ash values i.e. *Peristrophe bicalyculata* (9.97 %) ,*Oldenlandia corymbosa* (10.07 %), *Justicia procumbens* (11.09 %) and *Rungia repens* (12.49 %).

Acid insoluble ash content

Acid insoluble ash content of *Fumaria parviflora* (1.07 %) along the material collected in different season does not show significant variation (Table-2) while the closest value to the substitute/adulterant in descending order is *Oldenlandia corymbosa* (1.14%), *Justicia procumbens* (0.98%), *Rungia repens* (0.98%), *Peristrophe bicalyculata* (0.55 %) and *Polycarpea corymbosa* (0.46 %).

Amongst all substitutes/adulterants of *Fumaria parviflora*, the *Polycarpea corymbosa* showed the closest value of total ash content which showed that the *P. corymbosa* was more close to *F. parviflora* as compared to other substitutes/adulterants of *F. parviflora.*

Alcohol soluble extractive

Alcohol soluble extractive value of *Fumaria parviflora* (14.39%) along the material collected in different season does not show significant variation (Table-2) while the closest value to the substitute/adulterant was of *Rungia repens* (13.92%). The values of *Justicia procumbens, Peristrophe bicalyculata, Oldenlandia corymbosa* and *Polycarpea corymbosa* was found to be 12.84%, 8.86% , 7.46% and 4.28% respectively.

Water soluble extractive

Water soluble extractive value of *Fumaria parviflora* (16.31 %) along the material collected in different season does not show significant variation (Table-2) while the closest value to the substitute/adulterant was of *Justicia procumbens* (17.34%), however the *Rungia repens* showed the maximum extraction (18.36%), while values of *Oldenlandia corymbosa*, *Polycarpea corymbosa* and *Peristrophe bicalyculata* was found to be 15.18 %, 15.15 % and 11.22 % respectively.

Chapter 4

4.a. *Bergenia ligulata* (Wall.) Engl. (Saxifragaceae)

Synonyms: ***Bergenia ciliata*** **(Haw.) Sternb.**

Sanskrit: Ashmaghna, Bhimayojini, Pashanabhedana, Shilabheda, Shveta.

Vernacular Names:

Assamese : Patharkuchi.

Bengali : Patharkuchi, Himasagara, Patrankur.

Gujrati : Pashanbheda, Pakhanbheda.

Hindi : Pakhanabheda, Silphara, Patharcua, Pakhanabhed, Silpbheda.

Kannada : Alepgaya, Pahanbhedi, Hittaga, Pasanaberu, Hittulaka.

Kashmiri : Pashanbhed.

Malayalam : Kallurvanchi, Kallurvanni, Kallorvanchi.

Marathi : Pashanbheda.

Oriya : Pasanbhedi, Pashanabheda.

Punjabi : Kachalu, Pashanbhed.

Tamil : Sirupilai.

Telugu : Kondapindi.

Distribution and habitat

A small perennial herb found throughout temperate Himalayas from Bhutan to Kashmir at an altitude between 2000-3000 m and in Khasia hills upto 1200 m altitude.

Morphological features.

Leaves variable coarsely hairy, sparsely hairy to glabrous, leaf apex obtusely pointed, alternate and exstipulate or with stipules adnate to the base of the petiole, or opposite and exstipulate. Flowers are usually hermaphrodite; sepals, petals and stamens symmetrically regular pinkish white. Calyx usually 5-numerous, more or less adnate to the ovary; lobes imbricate or valvate. Petals 5 or 4 (rarely 0), usually perigynous, often small imbricate or valvate. Stamens inserted with the petals, equaling or double their number, rarely indefinite. Ovary of 2 or 3-5 united carpel's, usually 2 or 3-5 celled with axile placentas, occasionally 1-celled with parietal placentas, ovules numerous, anatropous, erect or pendulous; styles as many as the carpels, free or more or less connate, stigma capitate, or lateral and subcapitate. Fruit capsular or baccate. Seeds usually numerous, usually albuminous.

Medicinal uses:

The plant is used for wound healing and ulcers, in vertigo, headache, dizziness , as antilithitic, in boils and blisters, in urinary calculi and other urinary diseases, as an antidiabetic, in heart diseases, haemorrhoids, stomach disorders and ophthalmia. The leaves are used as anti-inflammatory and for wound healing , leaves and shoot are used for wound healing and as haemostatic, for dissolving kidney stones. The rhizome is used as tonic, antipyretic, antidiarrhoeal, in ophthalmia, kidney stones, as analgesic, antilithitic, antipyretic, in myalgia and urinary complaints. The root is used in abdominal disorders, post partum haemorrhage, urinary calculi and other urinary disorders, haemorrhoids, heart diseases, as antidiabetic ,antipyretic, antidiarrhoeal, tonic, in ophthalmic diseases, skin diseases. pulmonary affections, as galactagogue, in toothache, dropsy, urinary troubles, as an anti-inflammatory, abortifacient, antipyretic, for wound healing, in menorrhagea, dysuria, urogenital disorders, as antilithitic and diuretic (Anon.2004). Acetone extract of root showed maximum decrease in the elevated blood glucose level and significant effect on the lipid profile (Singh *et al*., 2011).

Previous Phytochemical reports

The rhizome contained gallic acid, tannic acid and glucose (Anon.1989), β -Sitosterol, bergenin and galloylated leucoanthocyanidin-4- (2-0-galloyl) glucoside. Flavonols-quercetin and kaempferol alongwith their 3-rhamnosides quercitrin and afzelin ; β-sitosterol and arbutin derivatives have reported in the leaves (Anon.1990).

Previous pharmacognostic reports

Only the T.S of the rhizome has been done (Anon.1989 & Anon.2004) but study of T.L.S and R.L.S is remaining to be done. So a detailed study is conducted on rhizome of the plant.

Materials and methods

The plant material has been collected from hilly areas of Dehradun, Uttarakhand. Phytochemical analysis of rhizome of the plant for their secondary metabolites such as alkaloids, flavonoids, phenolics, steroids, quinones, mucilages and other compounds were done by standard methods of chromatography and spectrophotometry described in Chapter 2. Pharmacognostic, HPTLC finger printing and physico-chemical studies were done by using standard methods described in Chapter 2.

Results

Phytochemistry

Rhizome

Along with reported gallic acids the rhizome also showed the presence of phenolic acids such as vanillic acid, syringic acid along with high concentration of *p*-Hydroxy benzoic acids. Mucilage amounted to 4.2 % consisting of rhamnose and glucose. The rhizome also showed the presence of unidentified alkaloids and steroids.

Pharmacognosy

Macroscopic characters (Fig.51)

The rhizome was cylindrical and woody. The outer surface yellowish brown in colour and showed longitudinal wrinkles and furrows and ridges. Fracture short to fibrous , Odour faint aromatic.

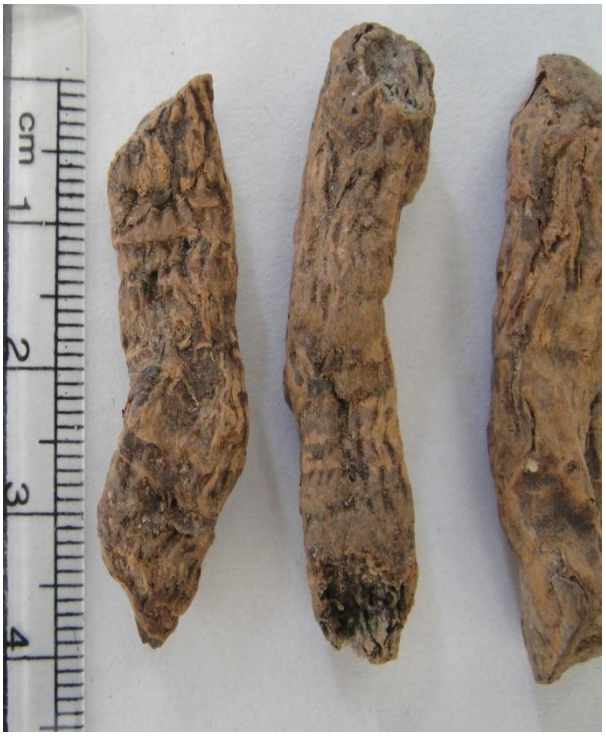


Fig.51. *Bergenia ligulata,* rhizome.

Microscopic characters

Rhizome : T.S (Fig.52)

The T.S of the rhizome was circular in outline with well developed cork and cortex. The outermost region of cork consisted of 3 to 5 rows of thick walled,

tangentially elongated cells containing starch grains and rosette crystals, of which the outer one or two rows of cells were ruptured and light brown in colour. The phellogen was a single row of narrow thin walled tangentially elongated cells followed by phelloderm consisted of 4 to 6 layers of thin walled tangentially elongated cells where in the 2 to 3 rows of cells towards cortex were polygonal in shape and filled with starch grains and rosette crystals. The wide secondary cortex consisted of circular to oval, thin walled parenchymatous cells with inter cellular spaces ,many of them were filled with rosette crystals (5-9µm) and starch grains. Starch grains were simple and of varying in shape, spherical and oval with blunt beak. some of the cortical cells were showed the deposition of light brown contents. Endoderm and pericycle were absent. Vascular bundles were 'V' shaped, open collateral, endarch and arranged in a ring. Both secondary xylem and phloem were not continuous but formed discrete bundles separated by broad medullary rays. The ray cells were thin walled and contained starch grains and rosette crystals while some of the cells showed the deposition of light brown contents. Phloem was 3-5 layered made up of usual elements. The xylem made up of fibres, tracheids, vessels and xylem parenchyma. The vessels were scattered and were simple and bordered pitted, some were spiral thickened. Central pith cells were similar to that of cortical parenchymatous cells and some of the cells showed the deposition of light brown contents.

Rhizome : T.L.S (Fig.53)

The cork cells were thick walled contained starch grains and rosette crystals. Parenchyma cells in the cortical region were large polygonal and contained starch grains and rosette crystals. Phloem cells appeared straight and upright. Tracheids were simple pitted. Vessels showed the presence of simple pits and few with spiral thickening also. .

Rhizome : R.L.S (Fig.54)

The thick walled cork cells followed by the large polygonal parenchyma showed the deposition of light brown contents. The pith parenchyma were square to rectangular in shape and the cell was filled with starch grains and rosette crystals.

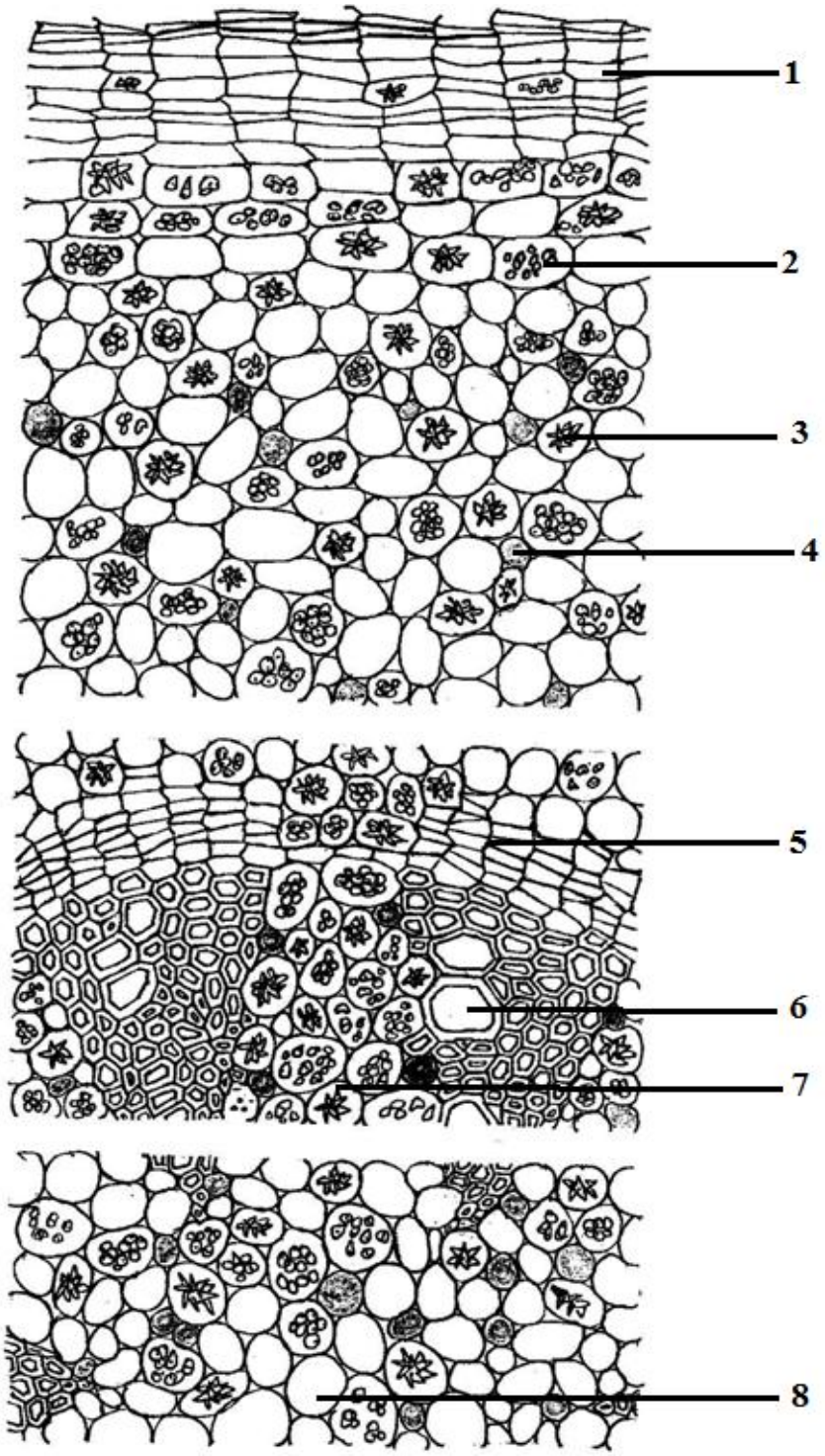


Fig.52. ***Bergenia ligulata*** **rhizome, T.S**: 1. Cork, 2. Parenchyma with starch grains, 3. Rosette crystal, 4. Parenchyma with light brown contents, 5. Phloem, 6. Vessels 7. Broad medullary rays, 8.Pith parenchyma.

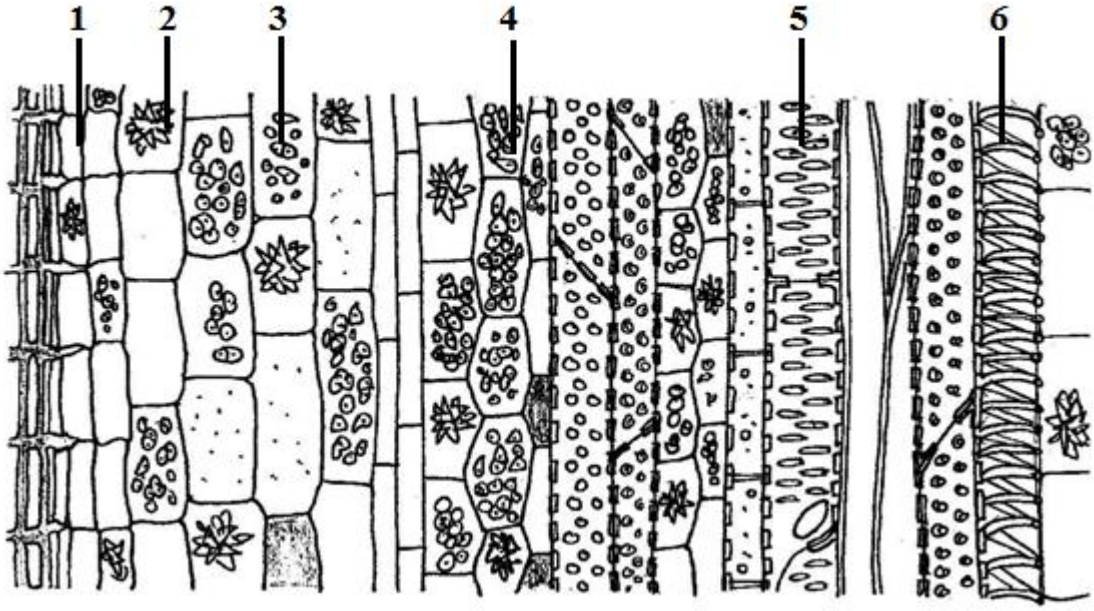


Fig.53. *Bergenia ligulata* rhizome, T.L.S:1. Cork, 2. Rosette crystal, 3. Starch grains, 4. Phloem rays, 5. Vessels with simple pits, 6. Spiral vessel.

Fig.54. *Bergenia ligulata* rhizome, R.L.S:1. Cork cells, 2. Parenchyma with light brown deposites, 3. Rosette crystal,4. Pith parenchyma with starch grains, 5. Pith parenchyma with with rosette crystals.

Rhizome : Powder study (Fig. 55)

The powder was characterized by the presence of groups of light brown thick walled cork cells, cortical cells showing deposition of light brown contents, groups of starch grains, rosette crystals, simple pitted vessels and spiral vessel.

Distinguishing features

Phytochemical markers

1. *P*-Hydroxy benzoic acid.
2. Vanillic acid.
3. Syringic acid.

Pharmacognostic markers

1. Light brown thick walled cork cells.
2. Parenchyma showing deposition of light brown contents.
3. Starch grains.
4. Rosette crystals.
5. Spiral and simple pitted vessels.

Physico-chemical analysis:

Table :8. Values obtained for the proximate analysis.

Sr.No.	Parameter	Mean ± SD (%)*			Average (%)
		Summer	Monsoon	Winter	
1.	Total Ash Content	12.03±0.31	12.14±0.11	12.09±0.12	12.09
2.	Acid Insoluble Ash content	1.18±0.31	1.21±0.30	1.18±0.26	1.19
3.	Alcohol soluble extractive	11.29±0.19	11.61±0.11	11.43±0.22	11.44
4.	Water soluble extractive	15.44±0.16	15.89±0.23	15.49±0.18	15.61

*Each value is a mean of 3 readings

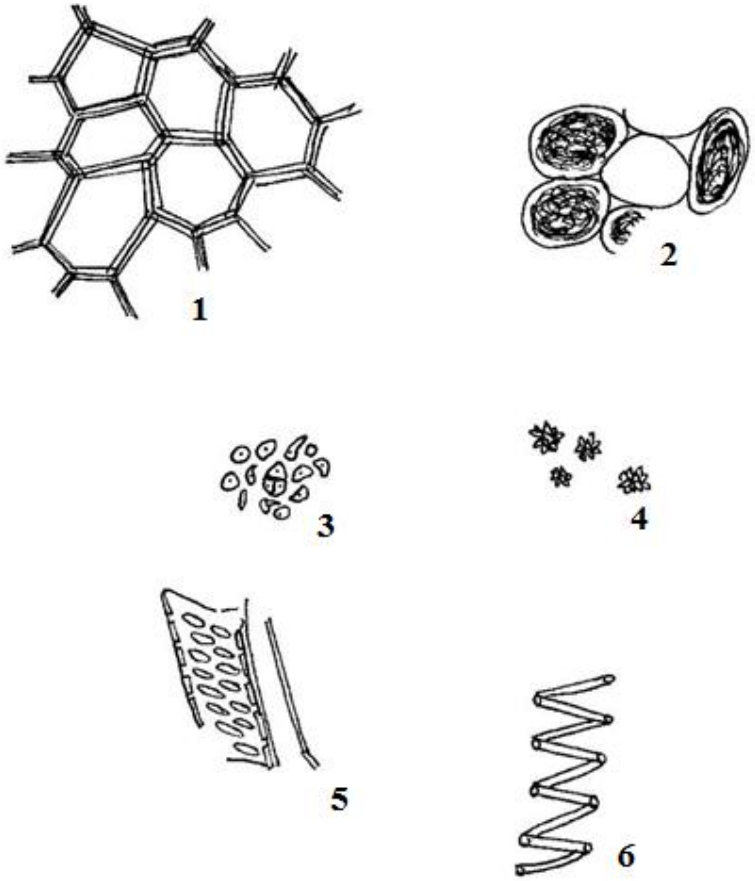


Fig.55. ***Bergenia ligulata*** **rhizome, powder study**:1. Cork, 2. Cortical cells with light brown deposition, 3. Starch grains,4. Rosette crystals, 5. Vessels with simple pits, 6. Spiral vessel.

4.b.*Aerua lanata* (Linn.) Juss.

Sanskrit: Astmabayda, Bhadra, Goraksaganja, Pasanabheda, Pashanabheda.

Vernacular names:

Bengali : Chaya.

English : Sunny Khur.

Gujarati : Gorakh-ganjo.

Hindi : Gorakh-ganja.

Kannada : Bilihindisoppu.

Malayalam : Cherula , Cherupula.

Marathi : Kapur-Madhura.

Oriya : Paunsia , Sanna sondo.

Punjab : Buikallan.

Tamil : Sirupoolai , Cerupulai.

Telugu : Pindiconda .

Distribution and habitat

The plant is a common weed found in all plains districts and upto 900 metres elevation. It is widespread in the drier parts of the tropics and subtropics of the Old World, Africa and Asia.

Morphological features.

The plant is an erect or prostrate herb with a long tap root, branched from near the base; branches many, terete, pubescent or woolly tomentose, striate. Leaves alternate, 2-2.5 by 1-1.5cm. on the main stem, 6-10 by 5-6 mm. on the branches, elliptic or obovate or sub-orbicular, obtuse or acute, entire, pubescent above, more or less white with cottony hair beneath; petioles 3-6 mm. long, often obscure. Flowers greenish-white, very small, sessile, often bisexual, in small dense subsessile axillary heads or spikes 6-13 mm. long, often closely crowded and forming globose clusters; bracteoles 1.25 mm. long, membranous, broadly ovate, concave, apiculate. Perianth 1.25-1.5 mm. long, sepals oblong, obtuse, sometimes apiculate, silky hair on the back; stigmas two in number. Utricle broadly ovoid, acute. Seed 0.85 mm. diameter, smooth and polished, black in colour

Medicinal uses:

The plant is anthelmintic and demulcent. It is used to treat malaria, skin diseases, indigestion and wounds (Daniel,2006). It is also used to treat diarrhea (Warrier *et al.*, 1994) .Extracts of the whole plant showed antibacterial, antifungal and cytotoxic activities (Chowdhury *et al.*,2002). The partially purified fraction of the petroleum ether extract of the plant reduced the development of solid tumour in mice significantly (Nevin and Vijayammal 2003). Leaf extracts increased urinary volume (Udupihille and Jiffry.1986) and showed antilithic property(Selvam *et al.*,2001). Various extracts of leaves were reported to inhibit angiotensin converting enzyme (Somanadhan *et al.*,1999). Alcoholic extract of shoots showed antidiabetic (Vetrichelvan and Jagadeeshan 2002), anti-inflammatory and diuretic activities in rats (Vetrichelvan *et al.*,2000) and produced a fall in blood pressure as well as negative chronotropic effect(Tripathi *et al.*,1985). Leaf paste is mixed with gingelly oil and given to treat piles. Leaf and root paste is applied to treat pimples and skin infections. Leaf decoction is given as anthelmintic and demulcent. Root and flower decoction is given to treat headache. Root decoction is used as an antidote for snakebite. Root powder is used as tooth paste to treat toothache (Retnam and Martin).

Previous Phytochemical reports

The plant is found to contain β-sitosterol (Aiyer *et al.*,1973, Aboutabl,1996, Chandra and Sastry 1990), α-amyrin (Aiyer *et al.*,1973,Chandra and Sastry 1990), hentriacontane (Chandra and Sastry 1990), campesterol (Aboutabl,1996,Chandra and Sastry 1990),stigmasterol (Aiyer *et al.*,1973, Aboutabl,1996, Chandra and Sastry 1990), stigmasterol acetate (Aboutabl,1996), daucosterol(Wassel and Amnar 1987), β-sitosterol palmitate (Aiyer *et al.*,1973), ergosterol, lupeol (Aboutabl,1996), β-amyrin (Chandra and Sastry 1990), olean-12-en-28-oic acid-3,16-dioxymethyl ester (Aboutabl,1996), kaempferol, kaempferol-3-galactoside, kaempferol–rhamnogalactoside (Afaq *et al*., 1991), starch (Afaq *et al*. 1991), free sugars (fructose, galactose, rhamnose and sucrose) (Afaq *et al*. 1991), alkaloids like canthin-6-one, β-carboline-1-propionic acid, 10-methoxy-canthin-6-one, 10-hydroxy-canthin-6-one, 10-O-β-glucopyranosyloxy canthin-6-one, 6-methoxy-β-carboline 1-propionic acid (Zapesochnaya *et al*., 1992), aervoside (Zapesochnaya *et al*., 1991,Zapesochnaya *et al*., 1992), aervolanine (Zapesochnaya *et al*., 1992),

flavonols like aervitrin (Zadorozhnii *et al.*, 1986), narcissi (Pervykh *et al.*, 1992) and a flavone chrysin (Zapesochnaya *et al*., 1992). The plant also shows the presence of saponins and phenolic acids such as vanillic and syringic acids (Mangalan 1988).

Previous pharmacognostic reports

Very little data available on the pharmacognosy of the root of this plant (Gupta *et al.*,2008)

Materials and methods

The plant material has been collected from Pavagadh, Vadodara,Gujarat. Phytochemical analysis of roots of the plant for their secondary metabolites such as alkaloids, flavonoids, phenolics, steroids, quinones, mucilages and other compounds were done by standard methods of chromatography and spectrophotometry described in Chapter 2. Pharmacognostic, HPTLC finger printing and physico-chemical studies were done by using standard methods described in Chapter 2.

Results

Phytochemistry

Root

There was no flavonoid in the root.The phenolic acids located were vanillic, syringic, ferulic (*cis* and *trans* isomers), melilotic, *p*- coumaric and *o*-coumaric acids. Mucilage amounted to 4.8% consisting of rhamnose and galactose . Steroids ,coumarins and saponins were also found to be present.

Pharmacognosy

Macroscopic characters (Fig.56)

The root was cylindrical and woody. The outer surface was yellowish brown in colour and showed longitudinal wrinkles and furrows and ridges. Fracture Short to fibrous , Odour faint aromatic.

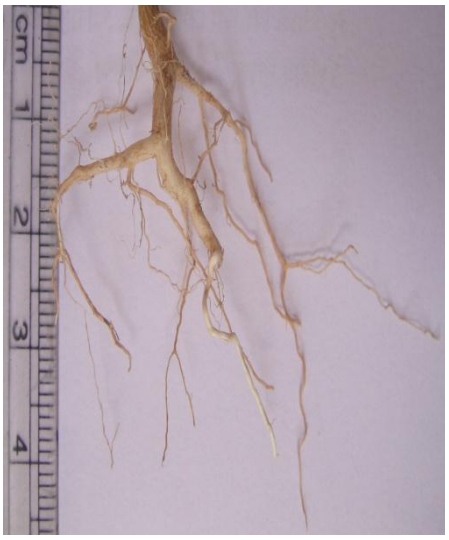


Fig. 56. *Aerua lanata* root.

Microscopic characters

Root : T.S (Fig.57)

The T.S of the root was circular in outline. The cork consisted of 3 to 6 rows of thin walled, tangentially elongated cells of which the outermost one or two rows of cells were slightly ruptured. Inner to the cork was the phellogen consisting of a single row of narrow thin walled tangentially elongated cells followed by one to three layers of phelloderm. The secondary cortex also was very narrow consisting of three to five rows of comparatively large polygonal or slightly tangentially elongated thin walled parenchyma cells, which were compactly arranged. Most of the cells were filled with rosette crystals and occasionally with rhomboidal crystals. The wood showed the secondary anomalous growth, consisting of a large number of vascular bundles arranged in successive rings separated by thin walled parenchyma filled with rosette and rhomboidal crystals. These vascular bundles were separated from the central phloem by thick walled parenchyma containing rosette and rhomboidal crystals. In the centre wood was a triarch. The phloem consisted of usual phloem

elements. Phloem rays were uni- to bi-seriate and the cells were thin walled and contained starch grains. Wood consisted of vessels, tracheids, fibres, wood parenchyma and rays. Xylem rays were thin walled containing starch grains in traces. Starch grains were small, spherical or ovoid in shape. Vessels were simple and bordered pitted. Spiral thickened vessels were also common.

Root : T.L.S (Fig.58)

Cork cells appeared rectangular with wavy walls. The cells of the cortex were thin walled, polygonal in shape and each cell contained rosette crystals and rhomboidal crystals. The fibres were thick walled and narrow lumened. Rays were spindle shaped biseriate and contained starch grains. The cells of the rays were thin walled, polygonal in shape and each cell contained 10-15 starch grains each. The bordered pits in vessels and tracheids were arranged loosely in 3-5 rows. The primary xylem vessel showed spiral thickening.

Root : R.L.S (Fig.59)

The phloem ray cells were thin walled and appeared hexagonal in shape. The xylem ray cells also were hexagonal in shape and filled with 5-10 starch grains each cell. The pits on the wall were of simple type.

Root : Powder study (Fig.60)

The components present in the powder were cork, rosette crystals, rhomboidal crystals, parenchyma, narrow lumened fibres, spiral vessles.

Distinguishing features

Phytochemical markers

1. Ferulic acid (*cis*- and *trans*-isomers).
2. *p*- Coumaric acid.
3. *o*-Coumaric acid.
4. Melilotic acid.
5. *p*-Hydroxy benzoic acid.

Pharmacognostic markers

1. Rosette crystals.
2. Rhomboidal crystals.
3. Spiral vessels.

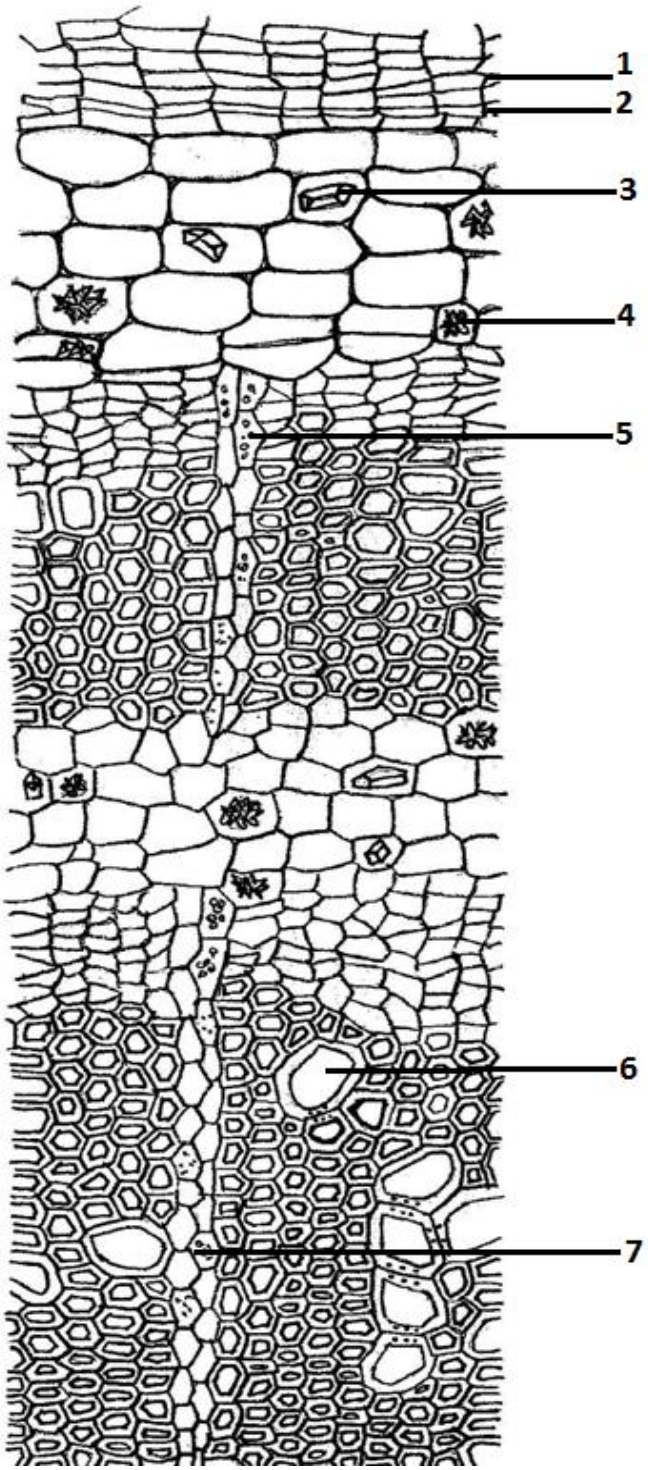


Fig.57. *Aerua lanata* root, T.S: 1. Cork, 2. Phellogen, 3. Parenchyma with rhomboidal crystal, 4. Rosette crystal, 5. Phloem ray with starch grains, 6. Vessels, 7. Xylem rays.

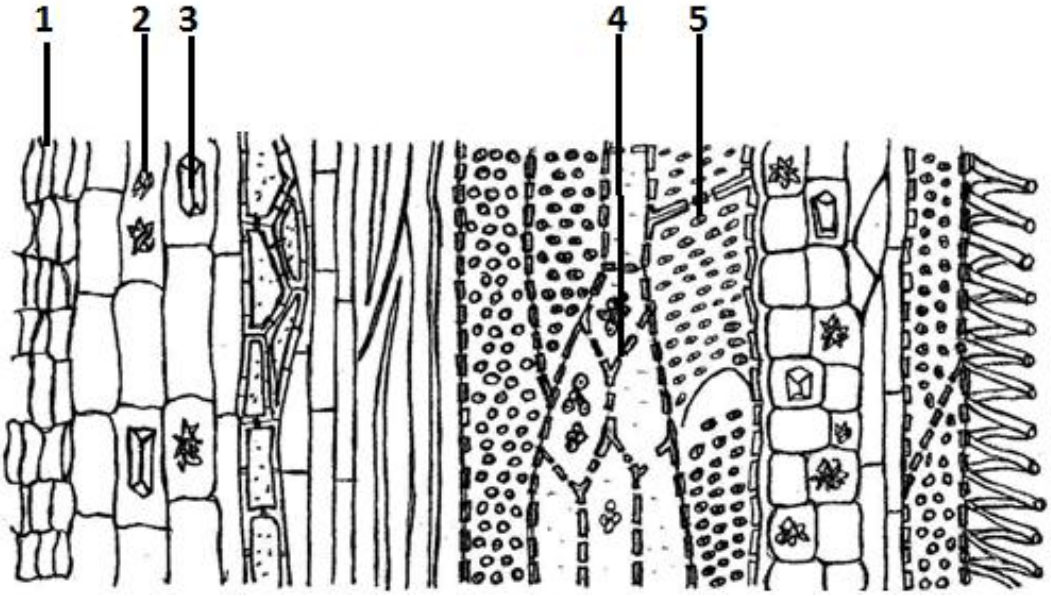


Fig.58. ***Aerua lanata*** **root, T.L.S**:1.Cork cells with wavy walls, 2.Rosette crystal, 3.Rhomboidal crystal, 4. Fibers with narrow lumen, 5. Spindle shaped medullary rays with starch grains, 6.Vessels with alternate bordered pits.

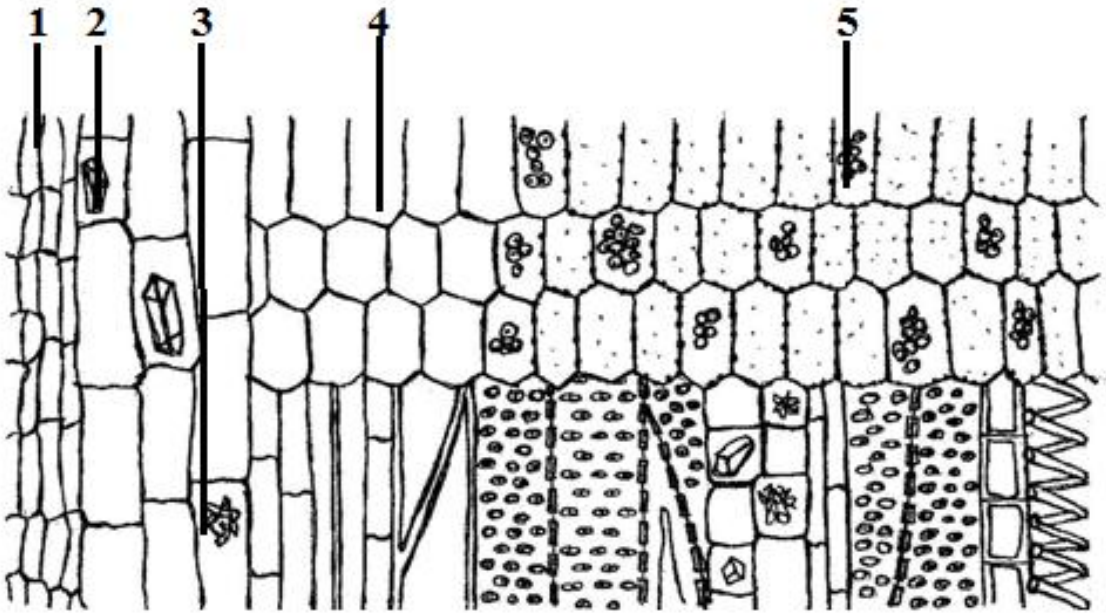


Fig.59. ***Aerua lanata*** **root, R.L.S**:1. Cork, 2. Rhomboidal crystal, 3. Rosette crystal, 4.Thin walled phloem rays, 5. Xylem rays with starch grains.

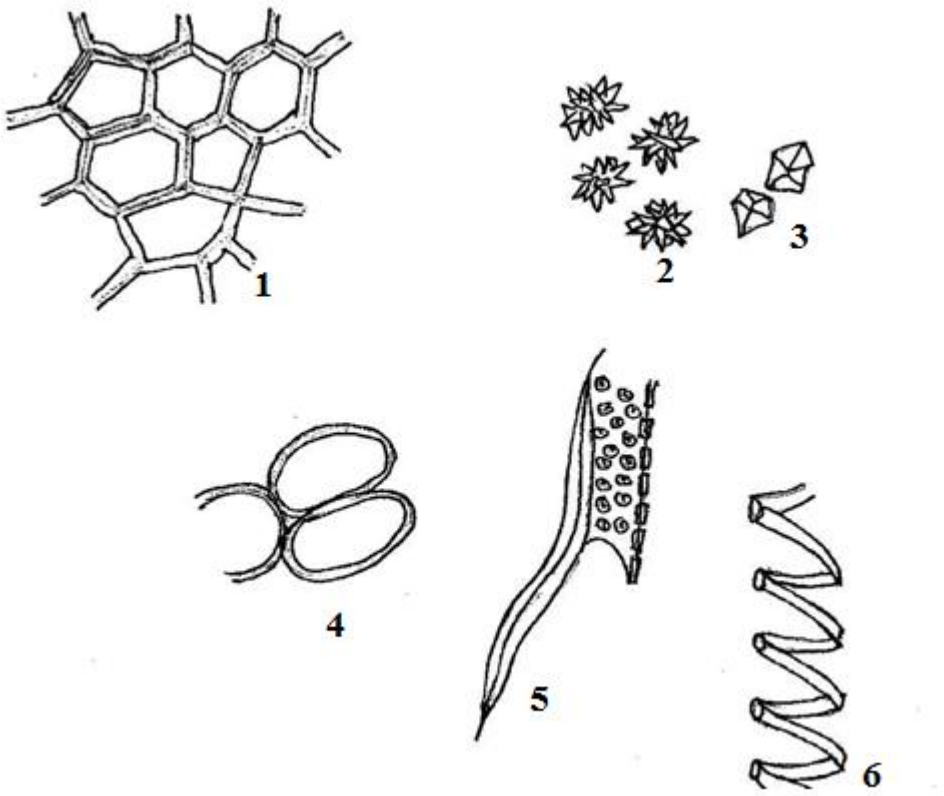


Fig.60. ***Aerua lanata*** **root Powder study:**1. Cork, 2. Rosette crystal, 3.Rhomboidal crystal, 4.Fragments of cortical parenchyma, 5. Fiber (adjoining vessel), 6. Spiral vessel.

Physico-chemical analysis:

Table :9 Values obtained for the proximate analysis.

Sr.No.	Parameter	Mean ± SD (%)*			Average (%)
		Summer	Monsoon	Winter	
1.	Total ash content	15.13±0.26	15.11±0.16	15.10±0.19	15.11
2.	Acid insoluble ash content	01.68±0.31	01.66±0.41	01.58±0.39	1.64
3.	Alcohol soluble extractive	4.16±0.16	4.13±0.19	4.17±0.22	4.15
4.	Water soluble extractive	10.13±0.36	10.32±0.29	10.28±0.37	10.24

*Each value is a mean of 3 readings

4.c.*Ammannia baccifera* Linn. (Lythraceae)

Synonyms: *Ammannia salicifolia* sensu Clarke

Sanskrit: Agnigarba, Brahmasoma, Davagni, Kurandika, Mahasyama, Pasanabheda.

Vernacular names:

Bengali : Dadmari, Banmarach.

English : Blistering Ammania.

Hindi : Dadamari, Mehudi, Jal Bhangro, Lal Babusi, Do-Patti-Ki-Kanduri, Lalbabusi.

Kannada : Agnivendrapaaku, Kaadugida, Kallurive.

Malayalam : Kallarvanchi, Kallur Vanchi, Kalluruvi, Nirummelneruppu.

Marathi : Bharajambhula, Aginbuti, Agyo, Dadmari.

Tamil : Kallarivi, Nirumelneruppu, Kalluruvipoondu,Tipputu.

Telugu : Agnivendapaku, Agnivendra-Paku, Agni Vendrapaku, Aginendramu.

Distribution and habitat

This species is globally distributed in the Paleotropics. Within India, it is found as a weed in rice-fields and marshy regions throughout.

Morphological features.

A glabrous, annual, branched herb reaching upto 60 cms. high with erect tetragonous stem. Leaves opposite, upto 6.6 cm, sessile, linear-oblong or oblong-lanceolate, subacute, much narrowed at the base. Flowers in dense axillary clusters short cymes, forming whorls in the axils; bracts filiform, shorter than the pedicels. Calyx tube hemispheric; teeth 4 (rarely 5), broadly triangular, acute; accessory teeth inconspicuous. Petals 0. Stamens 4, inserted in the middle of the calyx-tube; filaments filiform. Ovary superior, 1 celled; ovules numerous; style filiform, exserted; stigma capitate. Capsule globose, red, irregularly circumcise above the middle. Seeds subhemispheric.

Medicinal uses

The plant is used as an anthelmintic (guinea worm disease), antipyretic (Joshi, 1991; Pareek, 1994); in rheumatism (Singh and Pandey, 1980; Husain and Siddiqui, 1987; Siddiqui and Husain, 1992) and for skin eruptions (Chetty *et al*.,1998). The leaves are used in rheumatic pains and skin diseases (Bhatnagar *et al*.,1973; Kapoor and Kapoor, 1980; Shah *et al*.,1981; Saxena and Vyas,1983; Das,1995); in blisters (Siddiqui and Husain, 1992); ringworm (Singh *et al*., 1989) and intermittent fever

(Chetty *et al*.,1998), These are also used as analgesic and antipyretic (Bhatnagar *et al.*,1973).

Previous Phytochemical reports

The aerial parts were found to contain lawsone (Saoji *et al*.,1972). The tannin content in the 50 per cent ethanolic extract of the plant was 11.63 per cent (Atal *et al*., 1978).The fruits were reported to contain hentriacontane, dotriacontanol, triacontanediol and β-sitosterol glucoside while the leaves contained ellagic acid and quercetin in addition to hentriacontane,dotriacontanol and β-sitosterol. Betulinic acid and lupeol were reported from the root (Thakkar *et al*.,1986).

Previous pharmacognostic reports

No pharmacognostic work have been done on root of this plant.

Materials and methods

The plant material has been collected from Vadodara,Gujarat. Phytochemical analysis of roots of the plant for their secondary metabolites such as alkaloids, flavonoids, phenolics, steroids, quinones, mucilages and other compounds were done by standard methods of chromatography and spectrophotometry described in Chapter 2. Pharmacognostic, HPTLC finger printing and physico-chemical studies were done by using standard methods described in Chapter 2.

Results

Phytochemistry

There was no flavonoid in the root. The phenolic acids located were vanillic, syringic, melilotic and gallic acids. Mucilage amounted to 7.5 % consisting of xylose. The root also showed the presence of alkaloid ephedrine and steroids while coumarins and saponins were found in good concentrations.

Pharmacognosy

Macroscopic characters (Fig.61)

The tap roots were vertical, slightly tortuous, smooth with many lateral wiry rootlets and gray in colour. Fracture short.

Fig.61. *Ammannia baccifera* root.

Microscopic characters

Root : T.S. (Fig.62)

The T.S of the root was circular in outline with well developed cork. The outermost region of cork consisted of 2 to 3 rows of thick walled, square and tangentially elongated cells, of which the outermost one or two rows of cells were ruptured and light brown in colour while inner 2 to 3 rows of cells were compressed and tangentially elongated. The Secondary cortex consisted of circular to oval, thin walled parenchymatous cells showing inter cellular spaces many of them were filled with rosette crystals (5-9μm). The root showed the anomalous secondary growth forming continuous ring of stele by secondary cambium which arises from the outer layers of collenchyma near the original epidermis. The cells of collenchyma were slightly oblong, compactly packed and contained rosette crystals. The central core of the root was occupied by a primary xylem. The phloem was few layered and contained parenchyma and sieve elements. Xylem consisted of fibres, tracheids, parenchyma and vessels. The vessels were simple, bordered pitted and scalariform thickened. The fibres were of both thin and thick walled. Medullary rays were narrow, mostly biseriate, thin walled and simple pitted. The central xylem portion was dominated by the vessels.

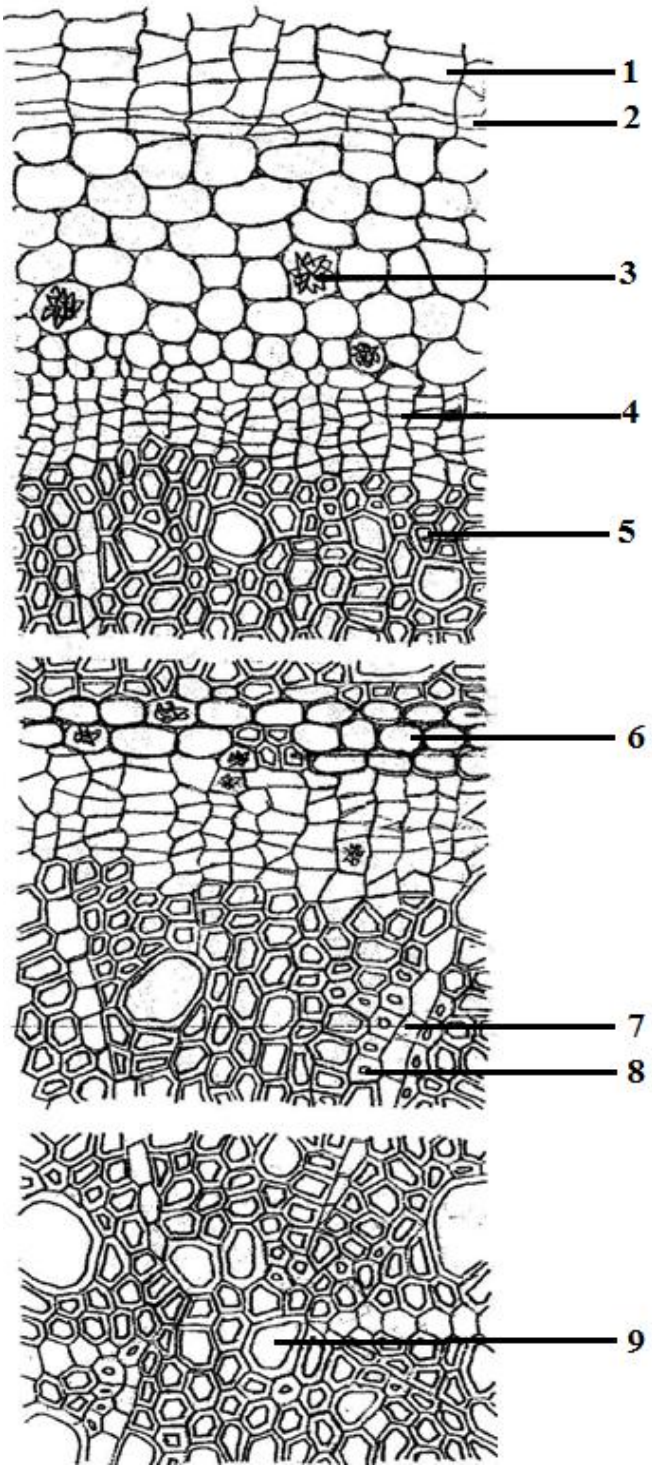


Fig.62. ***Ammannia baccifera*** **root, T.S:** 1. Cork, 2. Phellogen, 3. Parenchyma with rosette crystals, 4. Phloem, 5. Xylem, 6. Collenchyma, 7. Xylem rays, 8. Wood fibres, 9.Vessel.

Root : T.L.S (Fig.63)

Cork cells were thick walled rectangular followed by layers of polygonal cortical cells containing rosette crystals. The vessels were simple pitted, with 3-5 rows of simple pits. Xylem rays were biseriate and the cells were polygonal, thin walled. Scalariform vessel was with straight end walls.

Root : R.L.S (Fig.64)

Cork cells were thick walled rectangular laid one above the other. The cortical cells were polygonal, thin walled and contained rosette crystals. The collenchyma cells were polygonal in shape and were thick walled. The xylem rays were thin walled and had simple pits on its walls.

Root : Powder study (Fig.65)

The components present in the powder were light brown coloured thick walled cork cells, rosette crystals, collenchyma with rosette crystals, thin walled cortical parenchyma, thick walled fibres with pointed ends and simple pitted vessels.

Distinguishing features

Phytochemical markers

1. Gallic acid.
2. Melilotic acids.
3. Xylose.
4. Ephedrine.
5. Absence of flavonoid.

Pharmacognostic markers

1. Light brown coloured thick walled cork cells.
2. Rosette crystals.
3. Collenchyma.
4. Thick walled fibres with pointed ends.

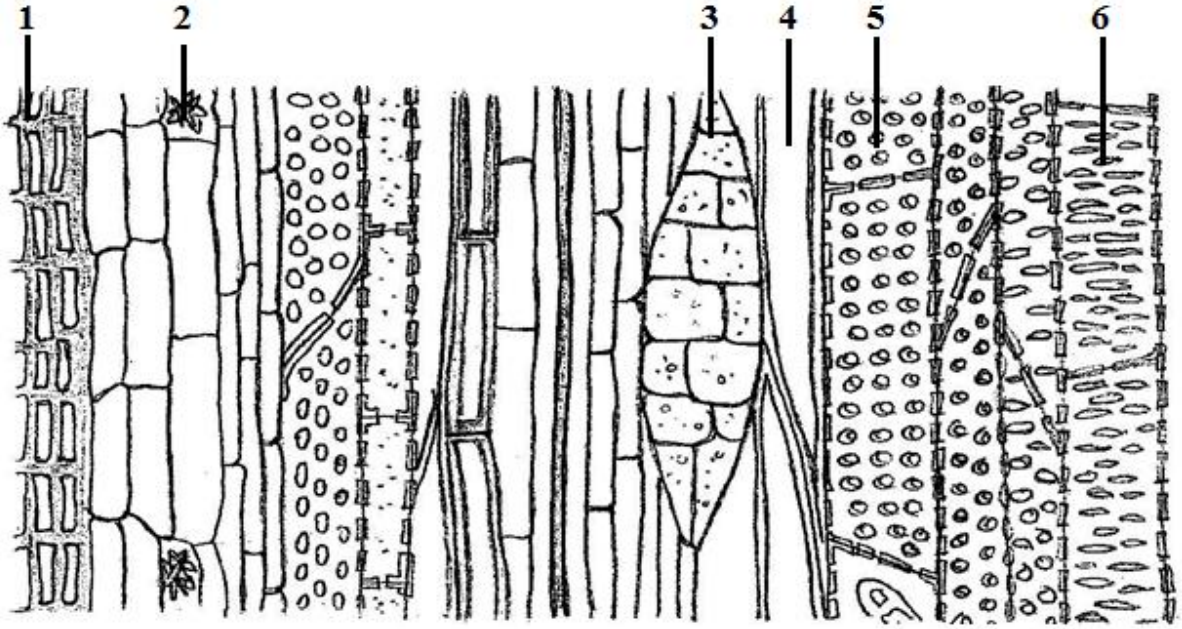


Fig.63. ***Ammannia baccifera*** **root, T.L.S:**1.Tthick walled cork cells with light brown walls, 2. Cortical parenchyma with rosette crystal, 3.Spindle shaped medullary rays, 4. Fibers, 5.Vessels with bordered pits, 6. Scalariform vessel with straight end walls.

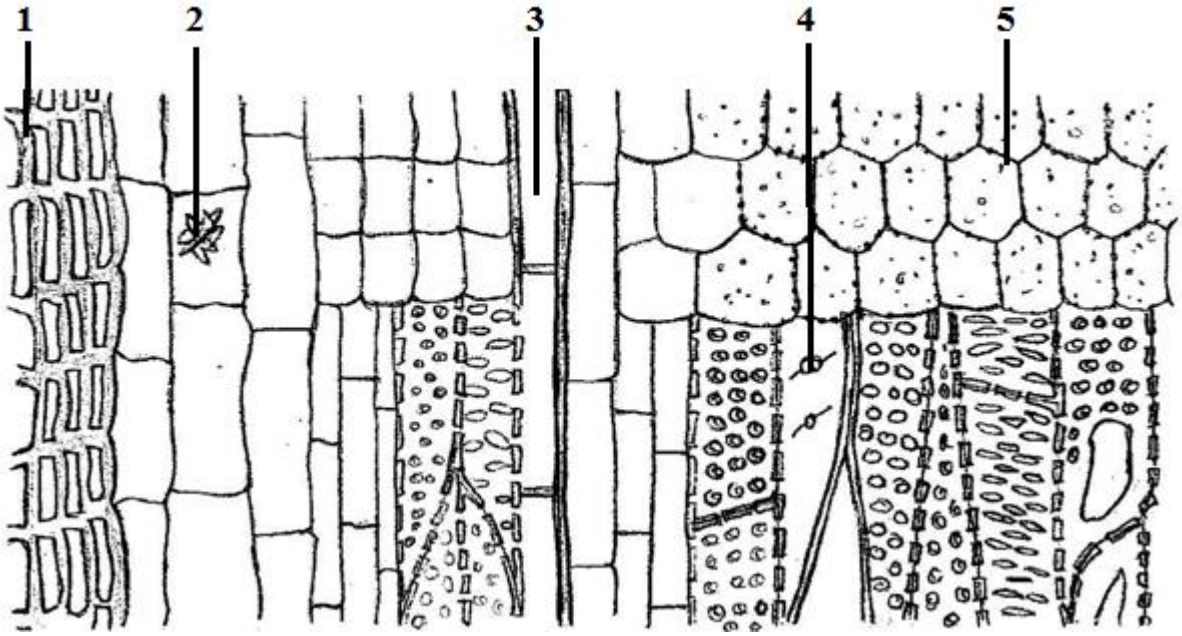


Fig.64. ***Ammannia baccifera*** **root, R.L.S**:1. Cork cells, 2. Rosette crystal, 3.Collenchyma, 4. Fibers with scanty pits, 5. Xylem rays.

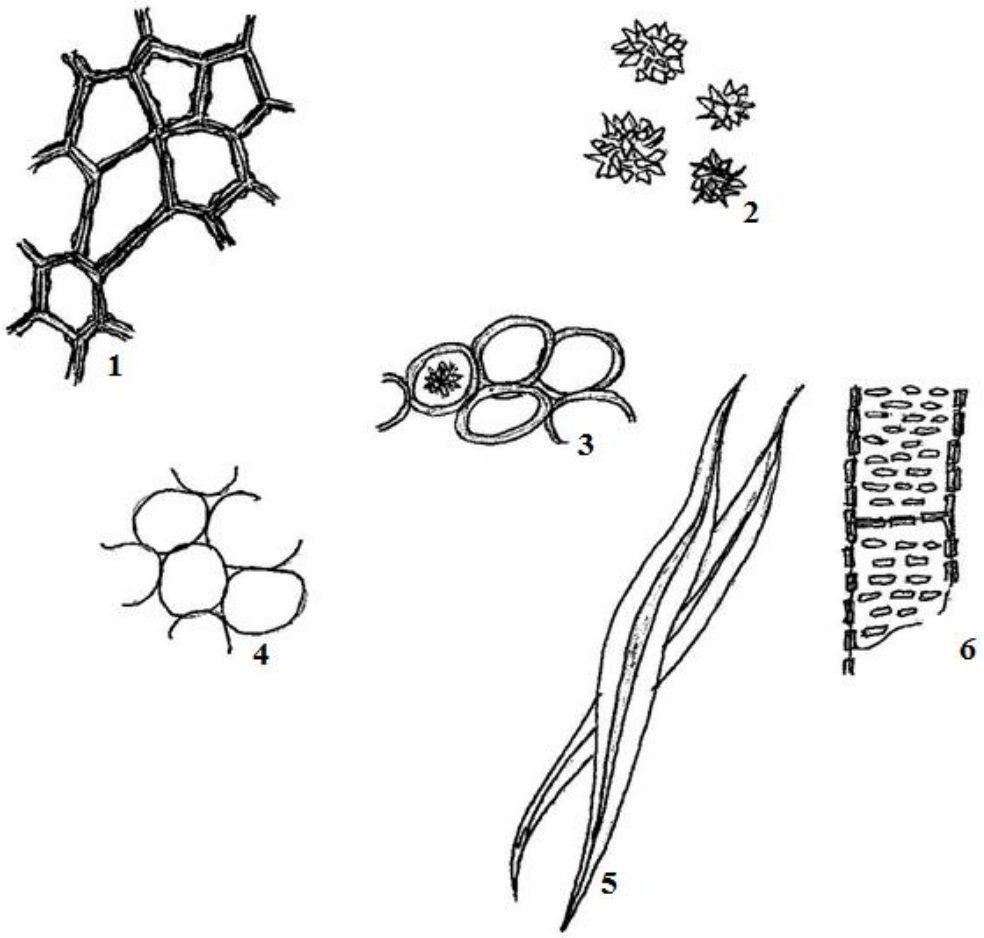


Fig.65. ***Ammannia baccifera*** **root, powder study:** 1.Light brown coloured thick walled cork cells, 2.Rosette crystals, 3. Collenchyma with rosette crystals, 4. Thin walled cortical parenchyma, 5. Thick walled fibres with pointed ends , 6.Simple pitted vessels.

Physico-chemical analysis:

Table 10:. Values obtained for the proximate analysis.

Sr.No.	Parameter	Mean ± SD (%)*			Average (%)
		Summer	Monsoon	Winter	
1.	Total ash content	05.29±0.13	05.21±0.19	05.14±0.18	5.21
2.	Acid insoluble ash content	0.77±0.25	0.69±0.26	0.68±0.19	0.71
3.	Alcohol soluble extractive	5.93±0.13	06.11±0.11	05.82±0.17	5.95
4.	Water soluble extractive	13.17±0.33	13.56±0.22	13.28±0.46	13.34

*Each value is a mean of 3 readings

4.d.*Celosia argentea* Linn.(Amaranthaceae)

Sanskrit: Sitivara, Vitunnaka, Sunishannaka, Indivara.

Vernacular names:

Assam : Boga kukurjoa.

Bengal : Swetmurga; Safed-morugphul,Swetmurgha.

English : Quail Grass, Silver-spiked,Cockscomb.

Gujarati : Lambadi, Lapadi.

Hind : Survali, Safed murga, Sufaid-murgha.

Kannada : Annesoppu,kanne hoo, karadoo.

Oriya : Gangachulia.

Punjab : Sarpankha, Sarwali.

Tamil : Pannakeerai, Pannai.

Telugu : Gurugu, Panchechettu, Gulugkura.

Distribution and habitat

An erect glabrous annual herb, 30 to 90 cm high, with conical to oblong feathery flowering spikes found commonly growing as a weed in cultivated fields throughout India upto an altitude of 1500 m.

Morphological features.

Annual herb, erect, 0.4-2 m, simple or with many ascending branches. Stem and branches strongly ridged and often sulcate, quite glabrous. Leaves lanceolate-oblong to narrowly linear, acute to obtuse, shortly mucronate with the excurrent midrib, glabrous; lamina of the leaves from the centre of the main stem 2-15 x 0.1-3.2 cm, tapering below into an indistinctly demarcated, slender petiole; upper and branch leaves smaller, markedly reducing; leaf axils often with small-leaved sterile shoots. Inflorescence a dense (rarely laxer below), many-flowered spike, 2.5-20 x 1.5-2.2 cm, silvery to pink, conical at first but becoming cylindrical in full flower, terminal on the stem and branches, on a long, sulcate peduncle up to c. 20 cm long, which often lengthens during flowering. Bracts and bracteoles lanceolate or the lower deltoid, 3-5 mm, hyaline, more or less aristate with the excurrent midrib, persistent after the fall of the flower. Perianth segments 6-10 mm, narrowly elliptic-oblong, acute to rather blunt, shortly mucronate with the excurrent midrib, with 2-4 lateral nerves ascending more than halfway up each segment, margins widely hyaline. Filaments very delicate, free part subequalling or exceeding the staminal

sheath, sinuses rounded with no or very minute intermediate teeth; anthers and filaments creamy to magenta. Stigmas 2-3, very short, the filiform style 5-7 mm long; ovary 4-8-ovulate. Capsule 3-4 mm, ovoid to almost globular. Seeds 1.25-1.5 mm, lenticular, black, shining, testa very finely reticulate.

Medicinal uses:

The leaves are used in poultices in China on infected sores, wounds and skin eruptions and in India mixed with honey on inflamed areas and painful afflictions such as buboes, abscesses etc. The whole plant is used as an antidote for snake bite and the root as a specific for colic, gonorrhoea and eczema. The water in which the leaves, flowers and stems have been boiled is used as a body wash for convalescents(Burkill, 1985).

Previous Phytochemical reports

Two rare isoflavones, 5-methoxy-6,7-methylenedioxy-2'-hydroxyisoflavone and 2',5-dimethoxy-6,7-methylenedioxyisoflavone, were isolated from the aerial parts of the plant(Jong and Hwang,1995). Alcoholic extract of seeds showed the presence of Celosian (Hase *et al.*,1996).

Previous pharmacognostic reports

Very little data available on the pharmacognosy of the root of this plant (Anon.2007).

Materials and methods

The plant material has been collected from Pavagadh, Vadodara,Gujarat. Phytochemical analysis of roots of the plant for their secondary metabolites such as alkaloids, flavonoids, phenolics, steroids, quinones, mucilages and other compounds were done by standard methods of chromatography and spectrophotometry described in Chapter 2. Pharmacognostic, HPTLC finger printing and physico-chemical studies were done by using standard methods described in Chapter 2.

Results

Phytochemistry

There was no flavonoid in the root. The phenolic acids located were vanillic, syringic and melilotic and *p*-cournaric acids. Mucilage amounted to 4.9 % consisting of xylose. The root also showed the presence of steroids while coumarins and saponins were found in good concentrations.

Pharmacognosy

Macroscopic characters (Fig.66)

The roots occurred in long, cylindrical somewhat tortuous and was branched with few lateral rootlets. The surface was grayish yellow to yellowish brown coloured and showed longitudinal fissures and transversely elongated wrinkles. The fracture was short.

Fig.66. *Celosia argentea* root

Microscopic characters

Root : T.S (Fig.67)

The T.S. of root was circular, showed the peripheral cork, wide central wood encircled by rings of xylem and phloem. The cork zone was well developed showed the outer 4-6 rows of compressed tangentially elongated cells. The walls of these cells were comparatively thick and light brown in colour. The cells of phelloderm were thin walled big polygonal and contained microspheroidal crystals. The secondary cortex was of 4-10 layers of somewhat broadly rectangular, thin walled parenchymatous cells, many of them contained microspheroidal crystals. The secondary growth was abnormal the primary cambium ceases the activity after some time to produce a central disc shaped xylem surrounded by phloem. Further secondary growth was on outer ring of cambium developed from the cortex. The behavior of this ring also was abnormal in that instead of producing a continuous ring of xylem or phloem it produces discrete secondary vascular bundles separated by 2 layered thick medullary

rays of rectangular cells. The pericycle showed isolated or small groups of fibers at intervals. The cortical cells between the central cylinder and outer ring of bundles also contained characteristic polygonal parenchyma cells containing microspheroidal crystals. Phloem was made up of usual elements. and also was contained yellow amorphous substances. The central primary xylem consisted of vessels, tracheids and fibers where the major portion was occupied by the broad vessels and distributed equally. The vessels were angular boarded pitted while the primary vessels showed annular and spiral thickenings. The scalariform vessels were also common. Xylem rays were indistinct.

Root : T.L.S (Fig. 68)

The cork cells were thick walled and light brown in colour followed by large polygonal cortical cells contained microspheroidal crystals. Pericyclic fibres were thick walled and narrow lumened. Phloem rays were thin walled. Xylem vessels had angular boarded pits. Primary xylem showed annular thickened vessel.

Root : R.L.S. (Fig. 69)

The cork cells were thick walled and light brown in colour followed by large polygonal cortical cells contained microspheroidal crystals. The ray cells present between two secondary vascular bundles were hexagonal in shape and contained microspheroidal crystals. The vessels were scalariform. Primary xylem showed spiral thickened vessel.

Root : Powder study (Fig.70)

The components present in the powder were thick walled brown colour cork cells, large polygonal shaped parenchyma contained microspheroidal crystals, angular boarded pitted,scalariform and annular thickened vessels.

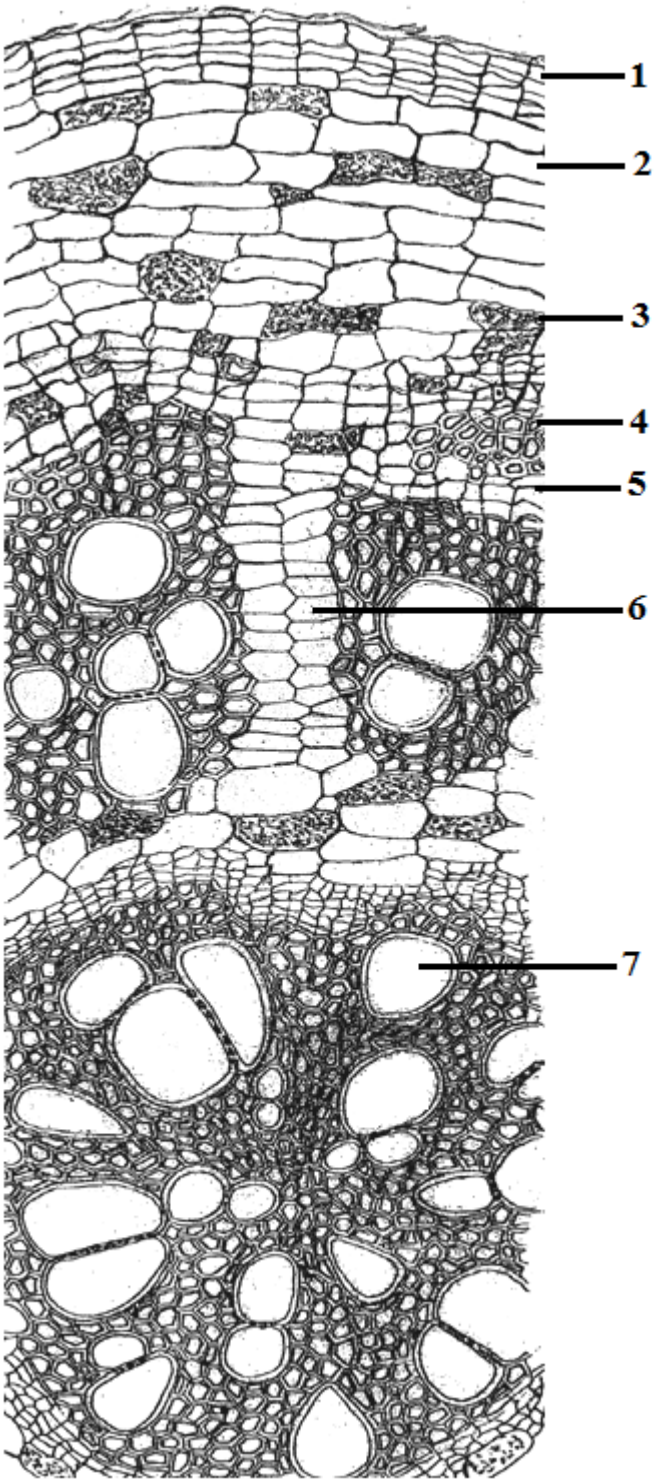


Fig.67.*Celosia argentea* root, T.S: 1. Cork, 2. large polygonal Parenchyma, 3.Microspheroidal crystals, 4. Pericyclic fibers, 5. Phloem, 6. Rays, 7.Vessels.

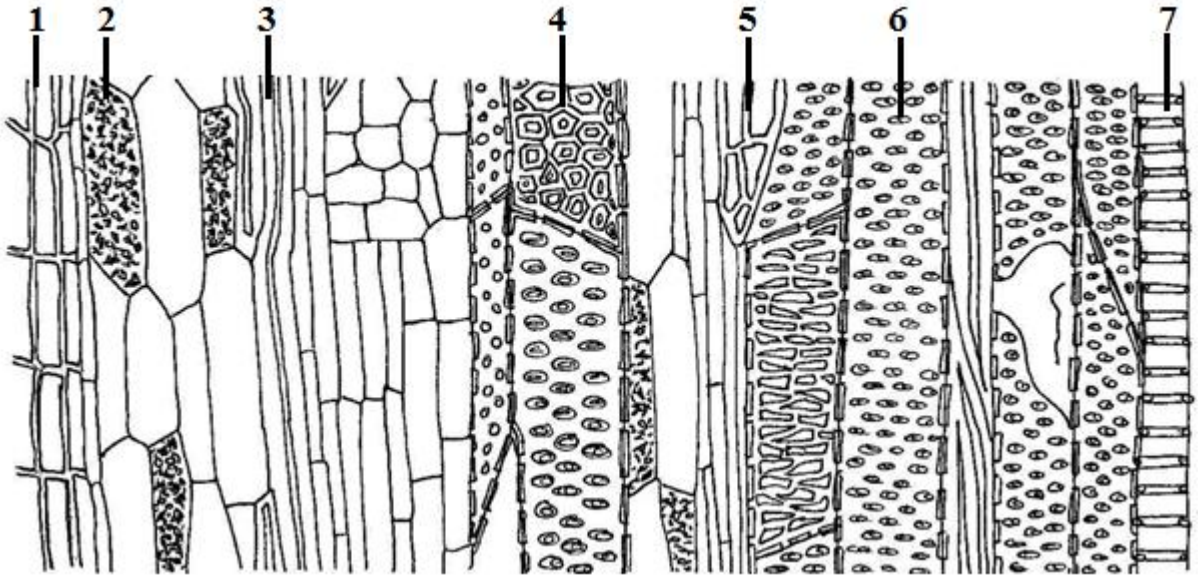


Fig. 68. ***Celosia argentea*** **root, T.L.S :** 1. Thick walled cork cells, 2.Parenchyma with microspheroidal crystals, 3. Pericyclic fibres,4. Vessel with angular boarded pits, 5. Narrow xylem rays, 6. Vessels with alternate bordered pits, 7. Annular thickened vessel.

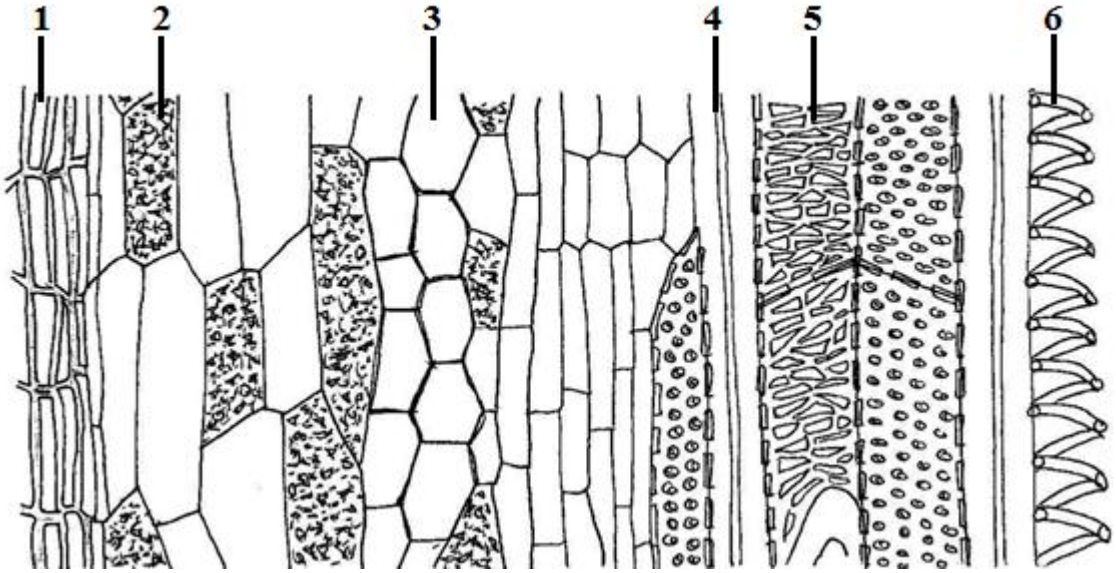


Fig.69. ***Celosia argentea*****root, R.L.S:**1. Cork cells, 2.Microspheroidal crystals, 3. Medullary rays, 4. Thick walled fiber, 5. Scalariform vessels, 6. Spiral vessel.

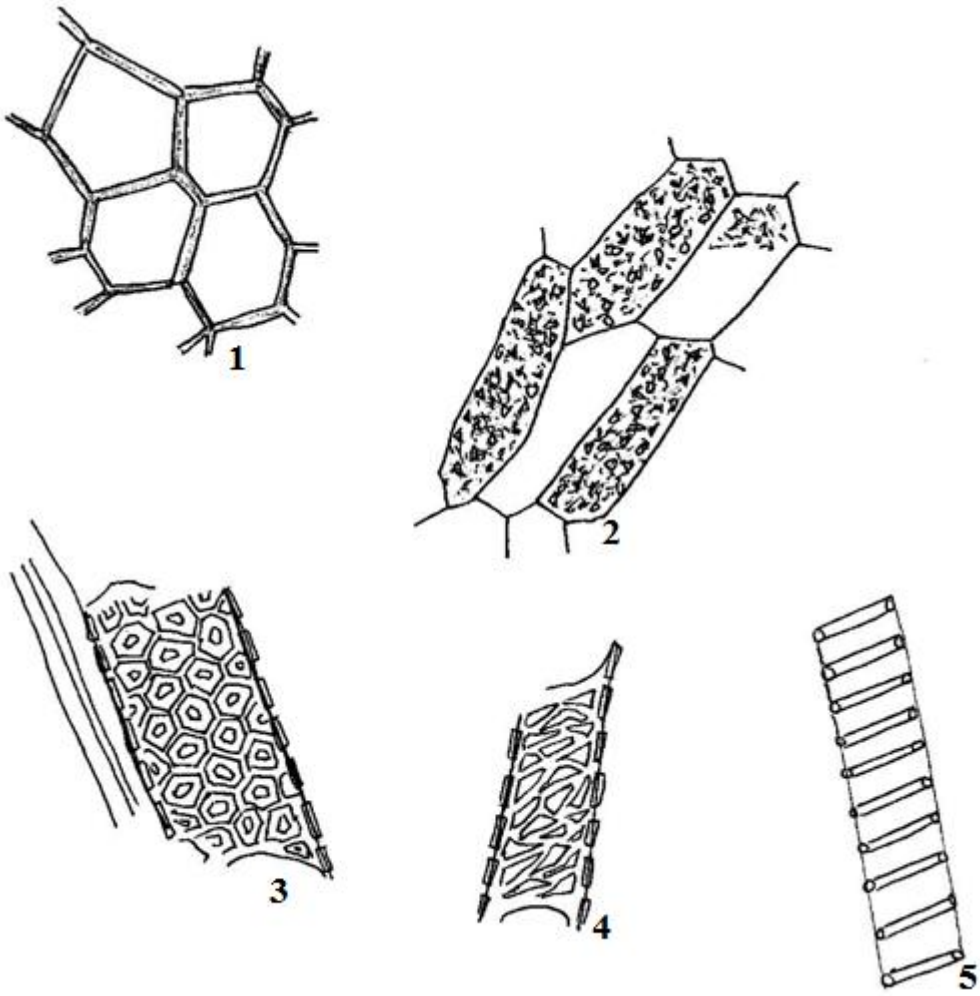


Fig.70. ***Celosia argentea*** **root, powder study:** 1. Cork cells, 2. Parenchyma with microspheroidal crystals, 3.Angular boarded pitted vessel, 4. Scalariform vessels 5. Annular thickened vessels.

Distinguishing features

Phytochemical markers

1. Melilotic acid.
2. *p*-Cournaric acid.
3. Xylose.
4. Absence of flavonoids

Pharmacognostic markers

1. Thick walled brown colour cork cells.
2. Microspheroidal crystals.
3. Angular boarded pitted vessels.
4. Scalariform vessels.
5. Annular thickened vessel

Physico-chemical analysis:

Table :11. Values obtained for the proximate analysis.

Sr.No.	Parameter	Mean ± SD (%)*			Average (%)
		Summer	Monsoon	Winter	
1.	Total ash content	4.33±0.42	4.46±0.51	4.32±0.49	4.37
2.	Acid insoluble ash content	0.93±0.11	0.94±0.09	0.93±0.09	0.93
3.	Alcohol soluble extractives	07.22±0.12	19.47±0.26	19.30±0.19	15.33
4.	Water soluble extractives	29.00±0.32	29.81±0.41	29.13±0.19	29.31

*Each value is a mean of 3 readings.

4.e.*Coleus amboinicus* lour. (Lamiaceae)

Synonyms: ***Plectranthus amboinicus*** **(Lour.) Spreng.** ;***Coleus aromaticus*** **Benth**

Sanskrit: Karpuravalli, Modayanti, Parnayavani,Pashanabheda, Pashanabhedi, Silabhedha.

Vernacular Names:

Bengali : Patherchur, Pathar Chur, Pashan Bhed.

English : Indian borage, Country borage.

Gujarati : Garmur ni bhaaji, Laanpadi, Lonpadi.

Hindi : Pathorchur, Amroda, Patharchur.

Kannada : Doddipatre.

Malayalam : Iribeli, Kannikkurka, Panikkurkaa.

Marathi : Karmelo.

Punjabi : Suravaali.

Tamil : Karpuravalli, Camparavalli, Omavalli, Muttainari, Ukkirikam.

Distribution and habitat

The plant is an aromatic,succulent perennial herb commonly cultivated in gardens throughout india and found wiled in Rajesthan.

Morphological features.

An erect annual, 3-10 dm high. Stem simple or branched, glabrous, strongly ribbed. Leaves 4-13 by 0.5- 5 cm, oblong-lanceolate or linear-lanceolate or rhombic or ovate, alternate, acute or acuminate at apex, petioles 1-3 cm long; leaf axils often with fulcate small leaves. Spikes usually solitary, pedunculate, cylindric with a conical, straw-coloured apex, sometimes tinged reddish, very dense, upto 10 cm long, 1-2 cm broad. Flowers perfect, the uppermost occasionally sterile, sessile; bracts and bracteoles 2.7 mm long, subequal, ovate, oblong, mucronate, pellucid, 1-nerved, perisistent. Perianth 6-10 mm long; lobes subequal, ovate, concave, mucronate, white or white with pink tip. Stamens 5, 3-5 mm long, united to form a 1.5-2 mm high cup, the free portion longer. Pseudo-staminodes minute, triangular or absent; anthers oblong. Ovary ellipsoid; style 1, 3-6 mm long, usually exceeding the perianth; stigma 2, minute. Fruit an utricle shorter than the perianth, 3-4 mm long, obovoid with rounded apex. Seeds dark reddish-brown, polished, shining.

Medicinal uses:

The plant used to treat malarial fever, hepatopathy, renal and vesical calculi, cough, chronic asthma, hiccough, bronchitis, helminthiasis, colic, convulsions, and epilepsy (Chopra *et al.*,1956, Kirtikar and Basu 1975, Nadkarni,1996).

Previous Phytochemical reports

The phytochemical study reveals the presence of various flavonoids like quercetin, apigenin, luteolin, salvigenin, genkwanin and volatile oil in the leaves (Rastogi and Mehrotra,1979). The main constituents are phenol, 2-methyl-5-(1-methylethyl), β-caryophyllene, γ-terpinene , α-bergamotene, m-cymene, α-caryophyllene, caryophyllene oxide and their isomers (Wang and Chen, 2005).

Previous pharmacognostic reports

Very little work has been done on roots of the plant (Hullatti and Bhattacharjee, 2011). In the present work it has been subjected for the detailed study. The root has been studied both for their phytochemical and pharmacognostic characteristics.

Materials and methods

The plant material has been collected from Pavagadh, Vadodara,Gujarat. Phytochemical analysis of roots of the plant for their secondary metabolites such as alkaloids, flavonoids, phenolics, steroids, quinones, mucilages and other compounds were done by standard methods of chromatography and spectrophotometry described in Chapter 2. Pharmacognostic, HPTLC finger printing and physico-chemical studies were done by using standard methods described in Chapter 2.

Results

Phytochemistry

The roots showed the presence of phenolic acids such as vanillic acid, syringic acid (in high concentration) along with melilotic and gallic acid . Mucilage amounted to 4.8 % consisting of xylose. The roots also showed the presence of unidentified alkaloids and steroids.

Pharmacognosy

Macroscopic characters (Fig.71)

Roots were cylindrical, vertical, slightly tortuous, with many lateral rootlets and gray in colour. Fracture short.

Fig. *Coleus amboinicus* root.

Root : T.S (Fig. 72)

The T.S. of root was circular, showed well developed peripheral cork and wide central wood encircled by few layered secondary cortex . The cork zone consisted of 6 to 13 rows of tangentially elongated thin walled cells, where the phellogen separates outer 6 to 8 layers of phellem from the inner 4 to 6 layers of phelloderm. The outer layers of the cork were broken at places and were thick walled. The phellogen was single layerd . The secondary cortex was a thin zone consisting of 5 to 6 rows of circular to oval shaped, thin walled cells, some of which showed the deposition of reddish brown contents. The cells were loosely arranged with inter-cellular spaces. The phloem was 8 to 10 layered thick made up of usual phloem elements. The phloem were capped by the sclerenchymatous fibers adjoining which

were stone cells. The sclerenchymatous fibers showed both broad and narrow lumen. The stone cells were thick walled and central lumen were unequal . The phloem rays were two celled wide, thick walled, usually tangentially elongated. The wood composed of many vessels, xylem parenchyma, fibres, tracheids and the medullary rays. The vessels were scalariform and bordered pitted. The fibres were thick walled, narrow lumened. Xylem rays were mostly biseriate and the cells were radialy elongated, thick walled with simple pits and few with starch grains. Scalariform vessel showed oblique end walls. The primary xylem showed mostly spirally thickened vessels.

Root : T.L.S (Fig. 73)

The cork cells were thick walled and rectangular in shape. Cortical parenchyma were polygonal in shape. The stone cells were thick walled with unequal lumen and were polygonal in shape. Xylem rays were mostly broad biseriate, spindle shaped wherein the cell walls were thick, pitted and few filled with starch grains.Vessels were scalariform.

Root : R.L.S (Fig.74)

The cortical parenchyma were polygonal in shape and showed the deposition of reddish brown contents. Sclereids were thick and thin walled. Phloem rays appeared rectangular and was elongated. The primary xylem region showed mostly spiral thickenings.

Root : Powder study (Fig. 75)

The components present in the powder were cork cells, fragments of cortical parenchyma with deposits of reddish brown contents, stone cells, ray parenchyma, scalariform vessel with oblique end walls and bordered pitted tracheids.

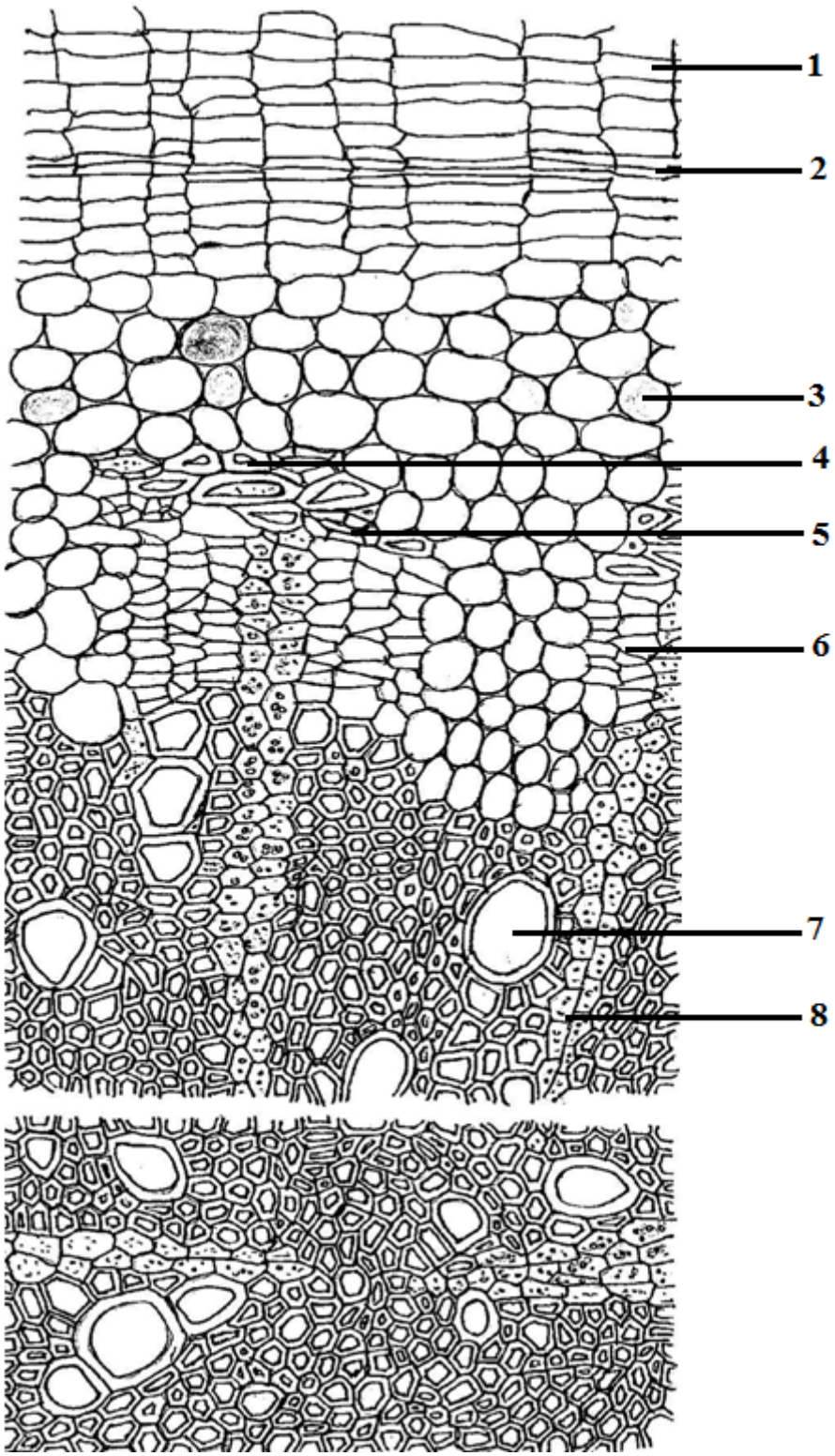


Fig.72.*Coleus amboinicus* root, T.S: 1. Cork, 2. Phellogen, 3. Parenchyma with deposits of reddish brown contents, 4. Sclereid, 5. Stone cell, 6. Phloem, 7.Vessel 8. Xylem rays.

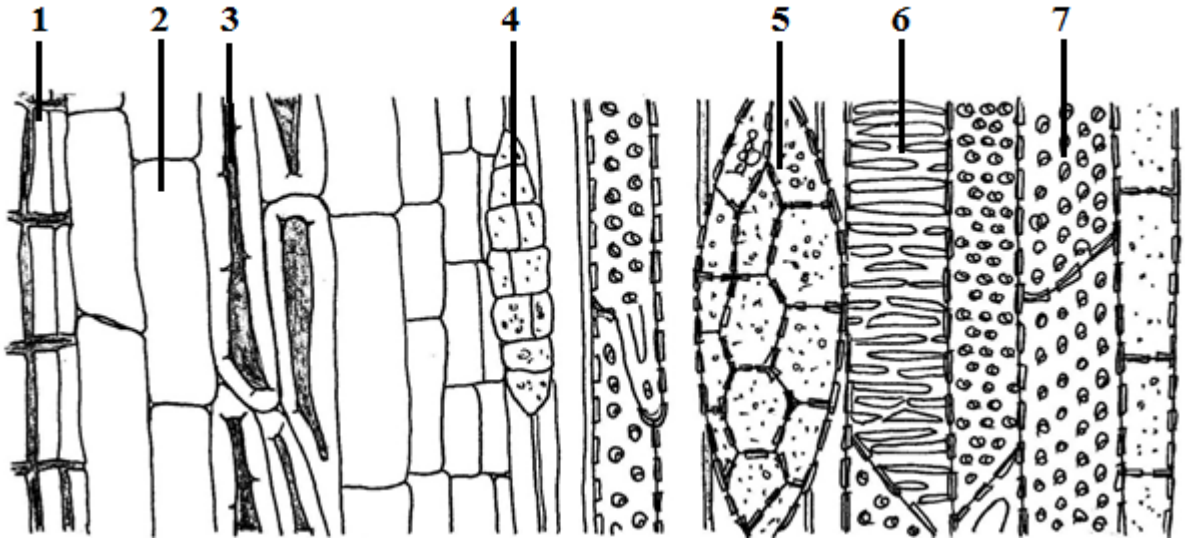


Fig.73.*Coleus amboinicus* root, T.L.S:1. Cork cells, 2. Cortical parenchyma, 3.Stone cell,4. Phloem ray with scanty pits, 5. Xylem rays, 6. Scalariform vessel,7.Bordered pitted vessel.

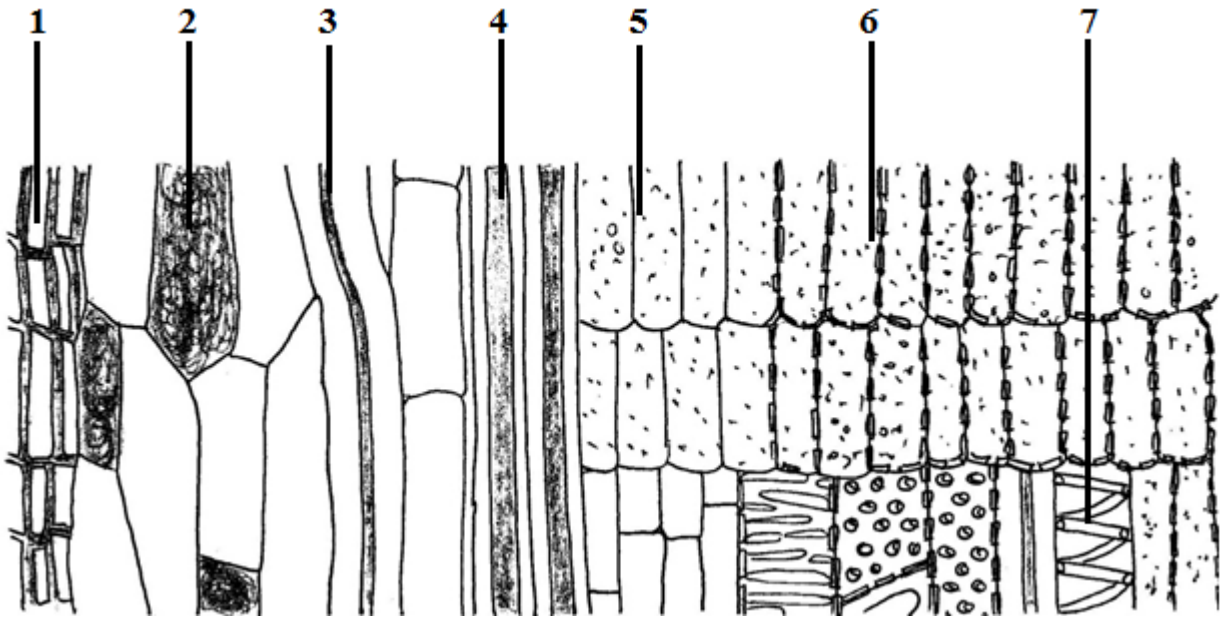


Fig.74.*Coleus amboinicus* root, R.L.S:1. Cork cells, 2. Cortical parenchyma with deposits of reddish brown contents, 3.Thick walled sclereid,4. Thin walled sclereid, 5. Phloem rays,6. Xylem rays, 7.Spiral vessel.

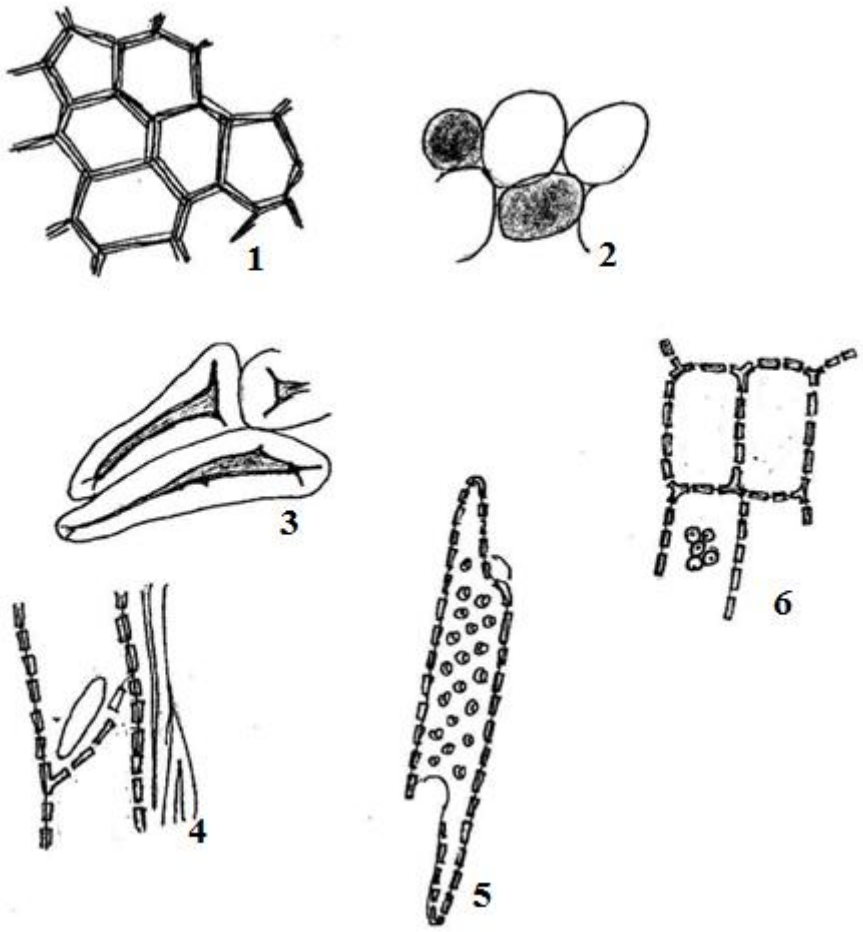


Fig.75. *Coleus amboinicus* root, powder study: 1. Cork, 2. Cortical parenchyma with deposits of reddish brown contents, 3. Stone cells, 4. Scalariform vessel with oblique end wall, 5.Bordered pitted tracheids, 6. Ray parenchyma.

Distinguishing features:

Pharmacognostic markers

1. Cortical parenchyma with the deposition of reddish brown contents.
2. Stone cells.
3. Thin and thick walled sclereids.
4. Scalariform vessel having oblique end walls.

Phytochemical markers

1. Gallic acid.
2. Vanillic acid.
3. Syringic acid.
4. Melilotic acid.
5. Xylose .

Physico-chemical analysis:

Table :**12.** Values obtained for the proximate analysis.

Sr.No.	Parameter	Mean ± SD (%)*			Average (%)
		Summer	Monsoon	Winter	
1.	Total ash content	08.93±0.21	08.99±0.12	08.96±0.18	8.96
2.	Acid insoluble ash content	0.97±0.09	0.99±0.10	0.98±0.08	0.98
3.	Alcohol soluble extractive	5.14±0.16	5.23±0.15	4.82±0.11	5.06
4.	Water soluble extractive	12.41±0.33	13..02±0.16	12.58±0.13	12.67

*Each value is a mean of 3 readings

4.f.*Glossocardia linearifolia* Cass.(Asteraceae)

Synonyms: ***Glossocardia bosvallia* (L.f.) DC.**

Sanskrit: Charak, Renu, Pithari.

Vernacular names:

English : Rock anethum.

Gujarati : Davanapada.

Hindi :Phattar-Suva, Seri.

Kannada : Parpataka.

Marathi : Phattar-Suva, Seri.

Tamil : Parapalanam.

Telugu : Parapalanamu.

Distribution and habitat

Found in Central India and Deccan Occuring in sandy and rocky tracts.

Morphological features.

The plant is small, prostrate or diffuse, tufted annual. Leaves bipinnatisect; segment narrowly linear. Heads small, yellow, heterogamous, about 8×8 mm. Achenes densely bearded,especially along the edge.

Medicinal uses:

The whole plant is used medicinally in the form of a confection, as an emmenagogue, in cases of suppressed menses, in doses of 1 to 4 drachms. It is useful also in fevers caused by *pitta* and vitiated *vayu* (Nadkarni,1954).

Previous Phytochemical reports

Root of the plant contains an essential oil; leaves, stems and flowers contain a bitter alkaloid (Nadkarni,1954).

Previous pharmacognostic reports

No work has been done on root of this plant. So the root of plant has been subjected for a detailed study.

Materials and methods

The plant material has been collected from Pavagadh, Vadodara,Gujarat. Phytochemical analysis of roots of the plant for their secondary metabolites such as alkaloids, flavonoids, phenolics, steroids, quinones, mucilages and other compounds were done by standard methods of chromatography and spectrophotometry described

in Chapter 2. Pharmacognostic, HPTLC finger printing and physico-chemical studies were done by using standard methods described in Chapter 2.

Results

Phytochemistry

The root of this plant showed the presence of flavone acacetin. The phenolic acids located were vanillic, syringic and ferulic (*cis* and *trans* isomers) acids. Mucilage amounted to 4.4 % consisted of xylose. The root also showed the presence of alkaloid and steroids.

Pharmacognosy

Macroscopic characters (Fig.76)

The roots were short, vertical, cylindrical somewhat tortuous and were with few lateral rootlets. The surface were grayish-yellow in colour. Fracture short.

Fig.76 *Glossocardia linearifolia* root

Microscopic characters (Fig.77)

Root : T.S (Fig.78)

The T.S of the root was circular in outline with a large central woody region and narrow cortex and outer bark. The cork consisted of 2 to 5 rows of cells wherein outer one or two rows were thick walled and inner, thin walled, light yellow coloured and tangentially elongated. The phellogen was indistinct. The secondary cortex was made up of polygonal or tangentially elongated thin walled parenchymatous cells. Some of them showed the deposition of reddish-brown contents. The central vascular

bundles were surrounded by discontinuous ring of pericycle. The pericyclic fibers were 3 to 4 cell wide and found associated with stone cells at periphery. Both pericyclic fibers and stone cells were thin and thick walled. The stone cells were mostly cubical to rectangular (180µm X35µm)with broad lumen, distinct prominent pits on their walls and without striations. Few stone cells with different shapes also present. The phloem composed of sieve elements and parenchyma , traversed by phloem rays; phloem rays 1-2 cells wide and were thin walled, isodiametric to slightly radially elongated. Wood consisted of fibres, tracheids, vessels and xylem parenchyma, traversed by xylem rays. Vessels were short (12-18µm) and mostly occurred singly with multiseriate simple and boarded pits. The tracheids were narrow (35 µm).The fibres were linear with blunt ends and broad lumen. Xylem rays uni to biseriate, thick-walled, cells radially elongated and pitted. The primary xylem showed the spiral vessels.

Root : T.L.S (Fig.79)

Cork cells appeared rectangular and walls were thick and wavy. The cells of the cortex were also appeared rectangular as of cork, but were larger in size. The stone cells showed distinct prominent pits on their walls. The phloem rays were thin walled. Xylem rays were fairly thick walled, oval and simple pitted. Tracheids contained 2 to 3 rows of bordered pits. The vessels had bordered pits and arranged alternately followed by broad lumen fibers.

Root : R.L.S (Fig.80)

Cork cells appeared rectangular with light yellow coloured, thick, wavy walls. The cortical parenchyma showed deposition of reddish-brown contents. The pericyclic fibers were narrow lumened. Xylem rays were thick walled, rectangular and simple pitted. The tracheids were narrow.

Root : Powder study (Fig.81)

The components present in the powder were cork with thick light yellow coloured wavy walls, cortical parenchyma with reddish-brown deposition, thin and thick walled stone cells, thick walled ray parenchyma with pits, blunt ends and broad lumen fibre, boarded pitted vessel adjoining tracheids.

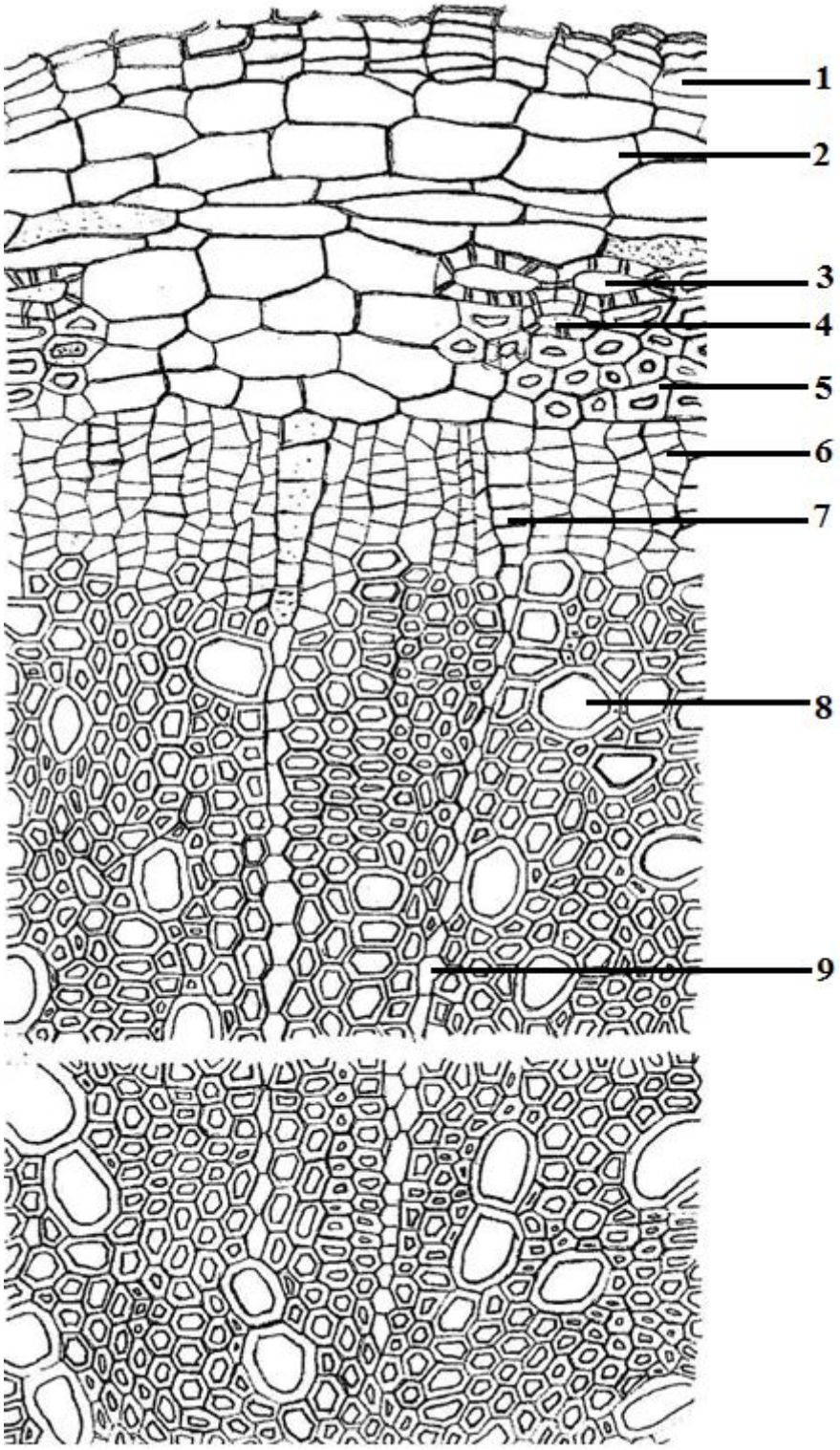


Fig.78.*Glossocardia linearifolia* root, T.S: 1. Cork, 2. Cortical parenchyma, 3.Stone cell, 4. Stone cell with narrow lumen, 5. Pericyclic fiber, 6. Phloem, 7. Phloem ray, 8. Vessel, 9. Xylem ray.

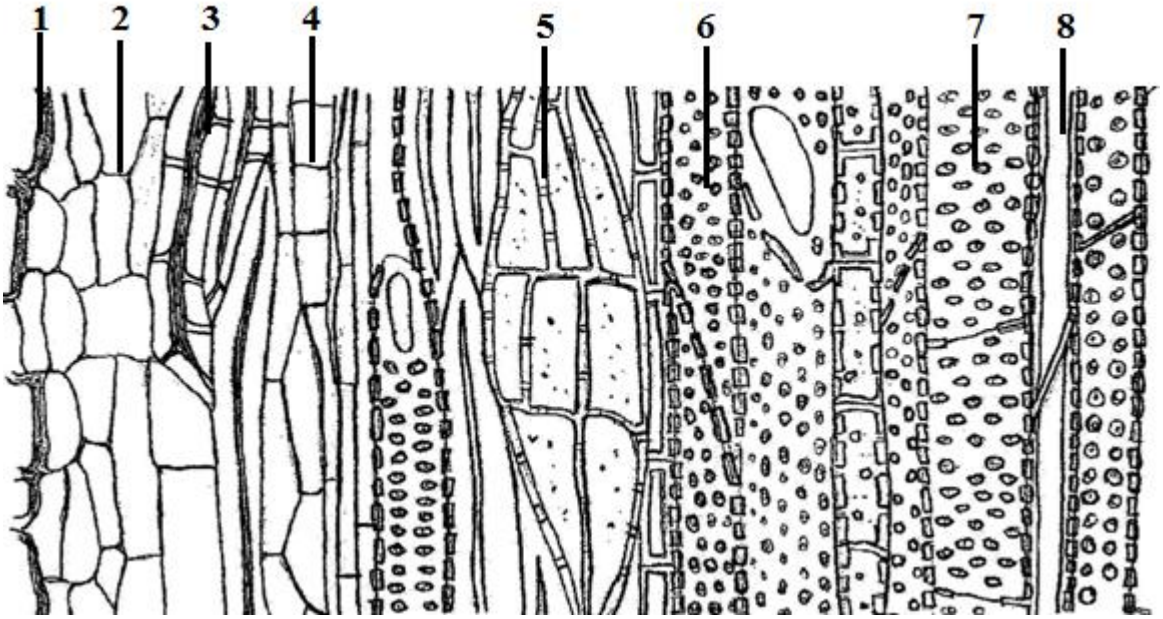


Fig.79. *Glossocardia linearifolia* root, T.L.S: 1. Cork, 2. Cortical parenchyma, 3. Stone cell with broad lumen, 4. Phloem ray, 5. Xylem rays, 6.Tracheid, 7. Bordered pitted vessels, 8.Broad lumen fiber.

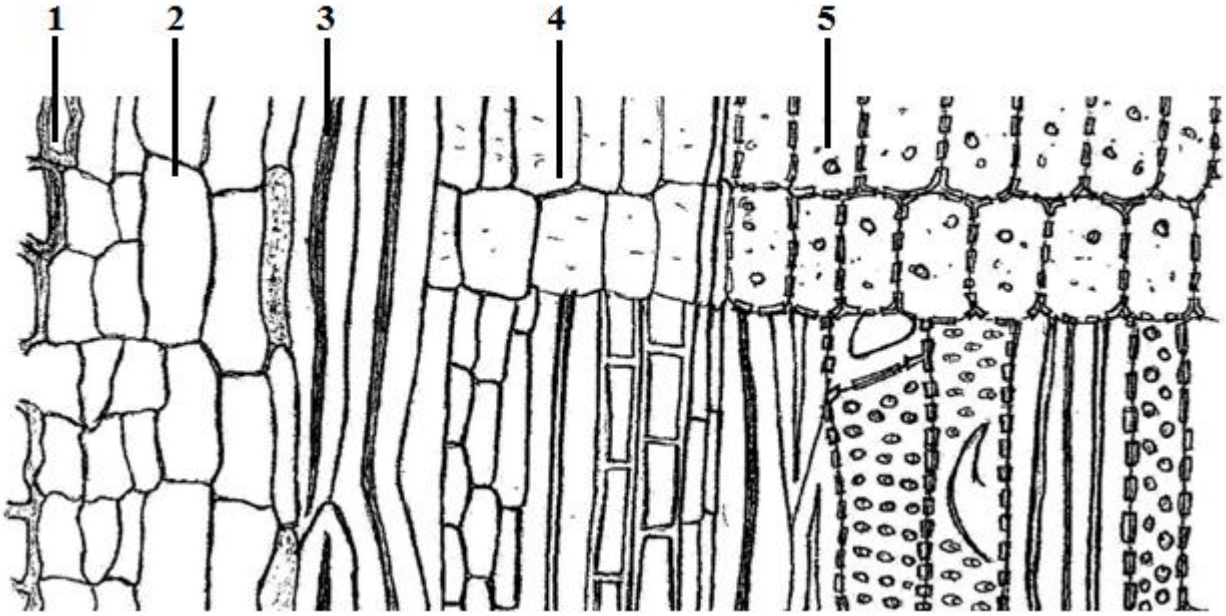


Fig.80. *Glossocardia linearifolia* root, R.L.S: 1. Cork, 2. Cortical parenchyma, 3. Pericyclic fiber, 4. Phloem ray, 5. Xylem ray.

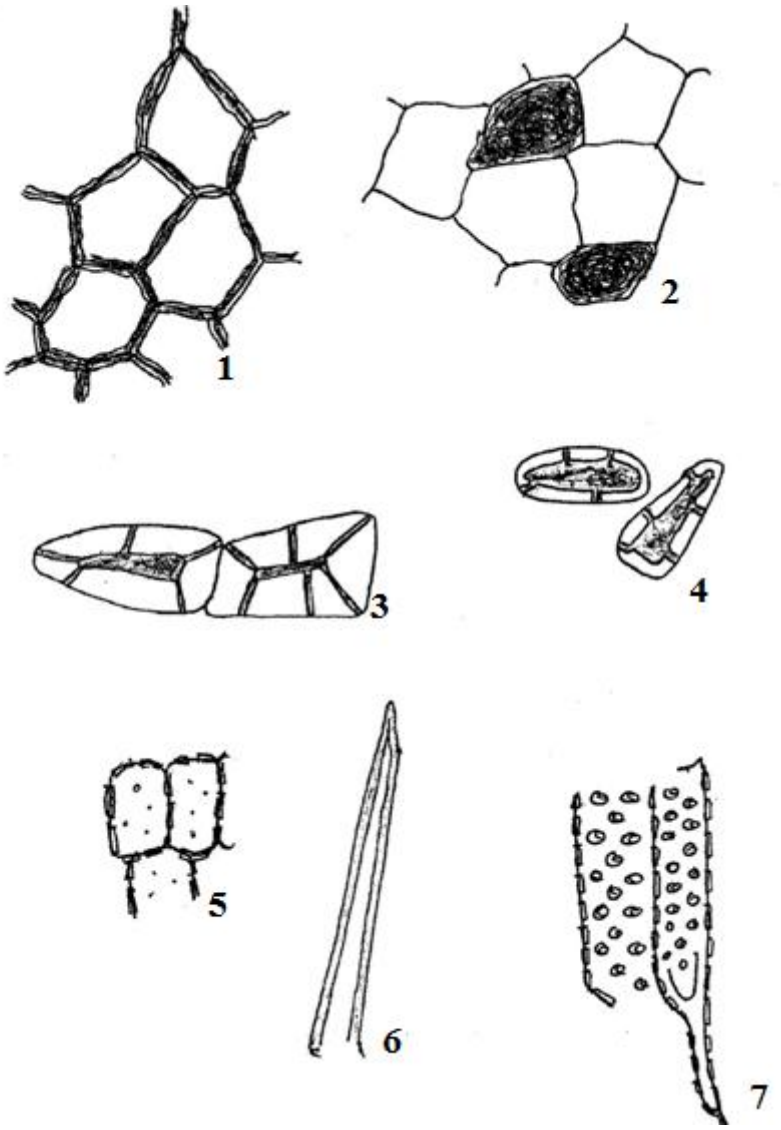


Fig.81.*Glossocardia linearifolia* root, powder study: 1.Cork with thick, light yellow coloured wavy walls, 2.Cortical parenchyma with reddish-brown deposition, 3. Thick walled stone cells, 4. Thin walled stone cells, 5. Thick walled ray parenchyma with pits, 6.Blunt ends and broad lumen fibre, 7.Boarded pitted vessel adjoining tracheids.

Distinguishing features

Phytochemical markers

1. Flavone acacetin.
2. Vanillic acid.
3. Syringic acid.
4. Ferulic (*cis* and *trans* isomers) acid.

Pharmacognostic markers

1. Cork with thick light yellow coloured wavy walled cork cells.
2. Cortical parenchyma with reddish-brown deposition.
3. Stone cells.
4. Thick walled ray parenchyma with pits.
5. Blunt ends and broad lumen fibres.

Physico-chemical analysis:

Table :**13** Values obtained for the proximate analysis.

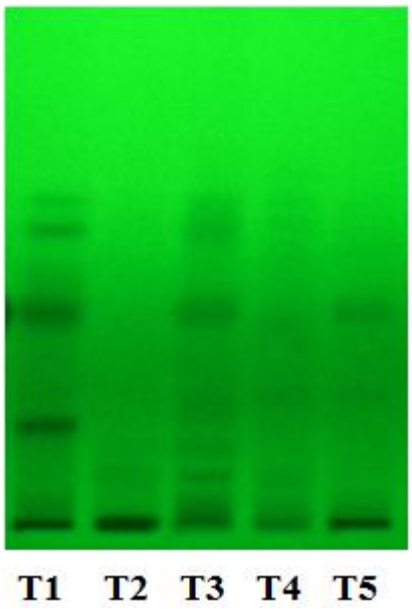
Sr.No.	Parameter	Mean ± SD (%)*			Average (%)
		Summer	Monsoon	Winter	
1.	Total ash content	06.33±0.18	06.48±0.11	06.16±0.28	6.32
2.	Acid insoluble ash content	0.57±0.10	0.59±0.13	0.57±0.09	0.58
3.	Alcohol soluble extractive	9.24±0.14	9.69±0.19	9.22±0.17	9.38
4.	Water soluble extractive	16.99±0.18	16.89±0.21	17.11±0.09	17.00

*Each value is a mean of 3 readings

4.g. HPTLC fingerprinting and Physo-chemical analysis of *Bergenia ligulata* and its substitutes/adulterants

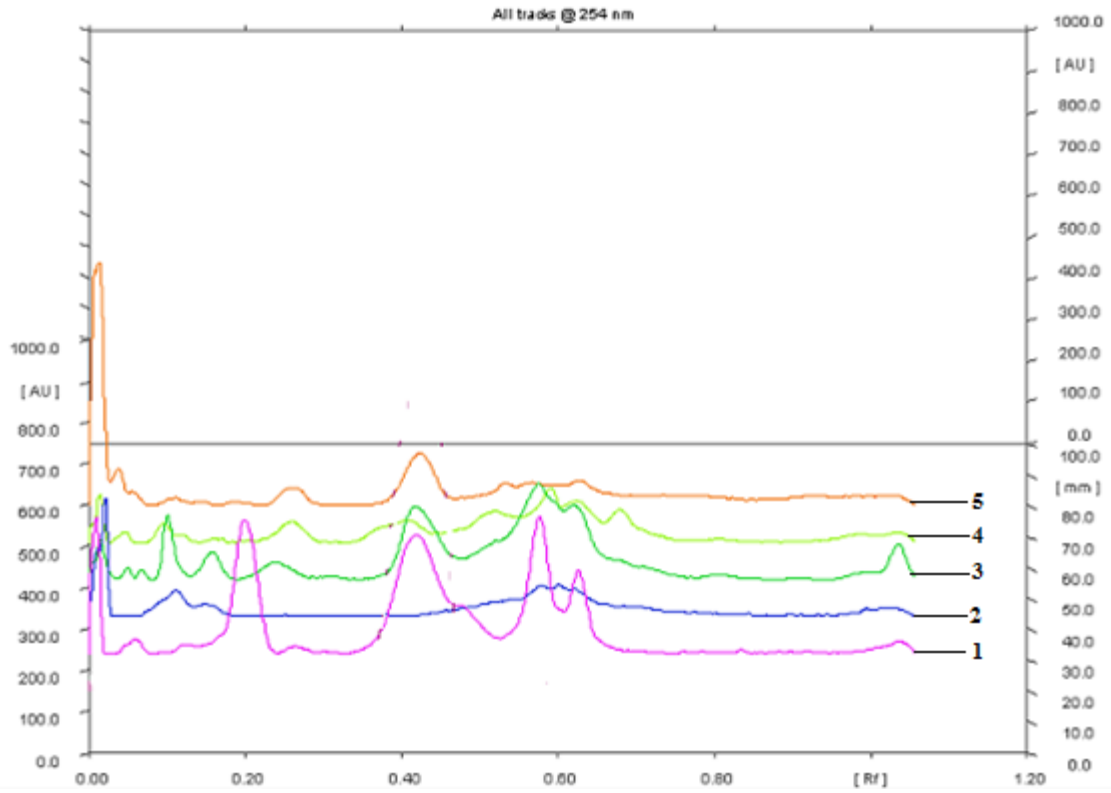
HPTLC fingerprinting

Figure 82.a : HPTLC chromatogram of *Bergenia ligulata* and its substitutes/adulterants (UV 254 nm).



(a).T1- *Bergenia ligulata,* T2- *Glossocardia linearifolia,* T3-*Coleus amboinicus,* T4-*Aerua lanata,* T5-*Ammannia baccifera*.

Figure 82.b : HPTLC chromatogram of *Bergenia ligulata* and its substitutes/adulterants (UV 254 nm).



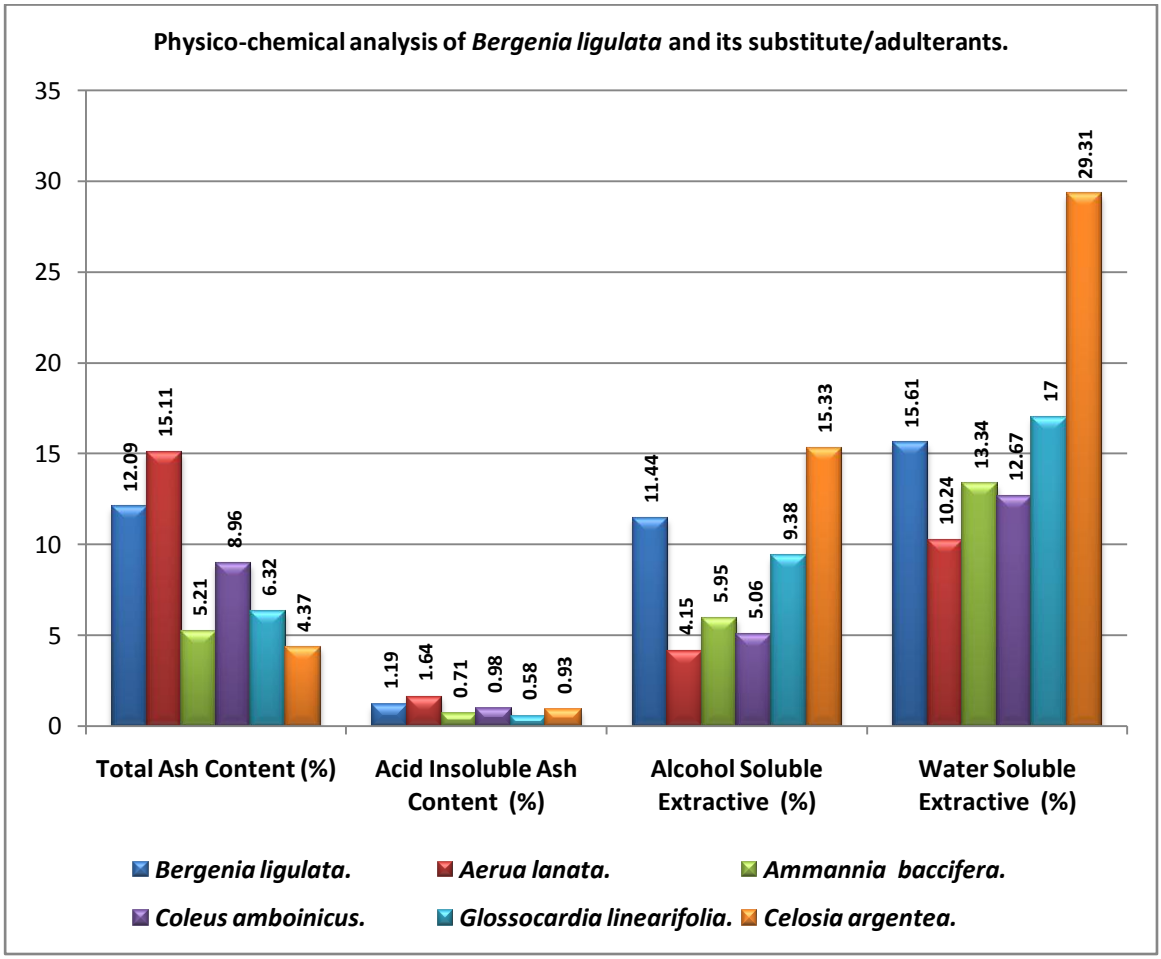
(b). 1- *Bergenia ligulata,* 2-*Glossocardia linearifolia,* 3-*Coleus amboinicus,* 4-*Aerua lanata,* 5-*Ammannia baccifera*.

HPTLC profile observed under UV 254 nm (figure82.b) the *Bergenia ligulata* showed the presence of 9 peaks and the major peaks were found at R_f 0.01, R_f 0.20, R_f 0.42, R_f 0.57 and R_f 0.63. The *Glossocardia linearifolia* also showed the presence of 9 peakes while *Coleus amboinicus, Aerua lanat* and *Ammannia baccifera* showed the 11,14 and 8 peaks respectively. *A. baccifera* was similar in 5 peaks and differed in 3 peaks *C. amboinicus* and *A. lanata* were found similar in 4 peaks but *C. amboinicus* differed in 7 peaks and *A. lanata* in 10 peaks while *G. linearifolia* was not show any peak similar to that of *B.ligulata* but differed in having 9 peaks.

HPTLC studies on *Celosia argentea* could not be conducted beacause of the great viscosity of the extract.

Physico-chemical analysis

Physico-chemical analysis of *Bergenia ligulata* and it substitutes/adulterants



Total ash content

Total Ash Content of *Bergenia ligulata* (12.09 %) along the material collected in different season does not show significant variation (Table-8) while the closest value to the substitute/adulterant in descending order is 15.11% (*Aerua lanata*), 8.96% (*Coleus amboinicus*), 6.32% (*Glossocardia linearifolia*), 5.21% (*Ammannia baccifera*) and 4.37% (*Celosia argentea*).

Acid insoluble ash content

Acid insoluble ash content of *Bergenia ligulata* (1.19 %) along the material collected in different season does not show significant variation (Table-8) while the closest value to the substitute/adulterant in descending order is 0.98%, 0.93%, 1.64%, 0.71% and 0.58% of *Coleus amboinicus* , *Celosia argentea*, *Aerua lanata*, *Ammannia baccifera*, and *Glossocardia linearifolia* respectively.

Amongst all substitutes/adulterants of *B. ligulata* , the *A. lanata* showed the closest value of total ash content which showed that the *A.lanata* was more close to *B. ligulata* as compared to other substitutes/adulterants of *B. ligulata.*

Alcohol soluble extractive

Alcohol soluble extractive value of *Bergenia ligulata* (11.44 %) along the material collected in different season does not show significant variation (Table-8) while the closest value to the substitute/adulterant was of *Glossocardia linearifolia* (9.38%), but the *Celosia argentea* showed the maximum extraction (15.33%) while values of *Ammannia baccifera, Coleus amboinicus* and *Aerua lanata* was found to be 5.95%,5.06% and 4.15% respectively.

Water soluble extractive

Water soluble extractive value of *Bergenia ligulata* (15.61 %) along the material collected in different season does not show significant variation (Table-8) while the closest value to the substitute/adulterant was of *Glossocardia linearifolia* (17.0%), but the *Celosia argentea* showed the maximum extraction (29.31%) while values of *Ammannia baccifera, Coleus amboinicus* and *Aerua lanata* was found to be 13.34%, 12.67% and 10.24 % respectively.

Amongst all substitutes/adulterants of *Bergenia ligulata* , the *C. argentea* showed the maximum extraction of phytoconstituents which reflect that the *C.argentea* could be chemically rich as compared to other substitutes/adulterants of *B. ligulata*.

Chapter 5

5.a.*Glycyrrhiza glabra* Linn (Fabaceae)

Synonyms : *Liquiritae officinalis* Moench.

Sanskrit names: Yasti, Yastimadhuka, Madhuka, Madhuyasti, Yastika, Yastyahva.

Vernacular names:
Assamese : Jesthimadhu, Yeshtmadhu.

Bengali : Yashtimadhu.

English : Liquorice root.

Gujrati : Jethimadha, Jethimard, Jethimadh.

Hindi : Mulethi, Mulathi, Muleti, Jethimadhu, Jethimadh.

Kannada : Jestamadu, Madhuka, Jyeshtamadhu, Atimadhura.

Kashmiri : Multhi.

Malayalam : Irattimadhuram.

Marathi : Jesthamadh.

Oriya : Jatimadhu, Jastimadhu.

Punjabi : Jethimadh, Mulathi.

Telugu : Atimadhuramu.

Urdu : Mulethi, Asl-us-sus.

Distribution and habitat

It is distributed in the Sub-tropical and warm temperate regions of the world, chiefly in the Mediteranean countries, South Europe, Asia Minor, Egypt, Turkistan, Iran, Siberia, Persia, Arab countries and Afganistan. In India, it is reported to be cultivated in Baramulla, Srinagar, Jammu, Dehradun, Delhi and South India.

Morphological features

The plant is an erect perennial shrub. Its principal or primary root does not generally grow deep but gives off a number of long tuberous secondary roots which may reach a length of four feet or more. The shoot system consists of an erect stem with a limited number of strong herbaceous branches which bear alternate odd pinnate leaves with five to seven pairs of ovate-oblong entire pale greenish leaflets. It also produces a number of long slender somewhat succulent stoloniferous under-ground branches (rhizomes) which spread out in all directions. and reach four to six feet in length.The flowers are medium sized, sessile, purplish-blue, or pale violet and

typically papilionaceous and the fruits are straight compressed or flattened oblong to linear echinate glandular pods one half to one and a half inches long, containing several kidney shaped seeds.

Medicinal uses

The roots are sweet, refrigerant, emetic in large doses, tonic, diuretic, demulcent,mild laxative, aphrodisiac, trichogenous, expectorant, emmenagogue, alexipharmic, haemostatic, alterant and intellect promoting. They are useful in hyperdipsia, cough, bronchitis, ulceration of urinary tract, retention of urine, gastralgia, gastric ulcer, cephalagia, fever, skin diseases, ophthalmic diseases, pharyngitis, haemorrhoids, consumption, hoarseness of voice, epilepsy, hiccough, erysipelas, anaemia, meno-metrorrhagia, intrinsic haemorrhage, hemicrania, urticaria. Decoction of root is good wash for falling and greying of hair. It is externally applied for cuts and wounds (Anon. 2005).

Previous phytochemical reports

Glycyrrhizine, prenylated biaurone, licoagrone; 7- acetoxy- 2- methyl-isoflavone, 7- methoxy- 2- methylisoflavone and 7- hydroxy- 2 methyl isoflavone; 4-methyl coumarin, liqcoumarin; isoflavone, glyzaglabrin (7,2'- dihydroxy 3',4'-methylenedihydroxy isoflavone); quercetin, quercetin-3- glucoside, kaempferol, astragalin, liquiritigenin and isoliquiritigenin (root). Other constituents reported include a flavanone rhamnoglucoside, chalcone glucosides, trans-isoliquiritigenin-4'-β-D-glucopyrano (isoliquiritin) and trans-isoliquiritigenin-4-ß-D-glucopyranoside (neoisofiquiritin); 7-hydroxy-4'-methoxyisoflavone (formetin),licuraside, liquiritoside, rhamnoliquiritin, triterpenoid, liquoric acid, 11-deoxoglycyrrhetic acid, liquiritic acid, isoglabrolide, glabrolide, deoxoglabrolide, glycyrrhizic acid, glycyrrhetol, 21α- hydroxy- 11- deoxyglycyrrhetic, and 24- hydroxyglycyrrhetic acids, 18α-ahydroxy glycyrrhetic acid, olean-12-en-3ß-o1-30 oic, olean- 11, 13 (18)-dien3ß-o1-30 oic acid, glabranine (5,7-dioxy-8-3 (3', 3'- dimethylallyl- flavanone), pinocembrin, prunetin, 4- hydroxy chalcone, liquiritigenin, licoflavonol (6- γ-γ-dimethylallylkaempferol), kuniatakenin, glycerol, licoricone, glabridirt, glabrol, liqurazid, liquiritin, 3-hydroxyglabrol, 4'-0-methyl glabridin, 3'-methoxyglabridin, glycyrrhetinic- acid; methyl olean-11,13 (l8)-diene-3, 24- dio1-30-oate, glabranine, forniononetin, glabrene, saponaretin (isovitexin), 24- hydroxy-11-deoxyglycyrrhetic acid, methyl olean 11, 13 (18) diene-3, glycerrhetol, 21α-hydroxy

isoglabrolide, licoflavonol, gly7arin, glyzaglabrin, licoisoflavones A, B and licoisoflavon, glycyrin, sugars and aspargin (root and other plant parts)(Anon.2005)

Previous pharmacognostic reports

Though T.S of root was described earlier (Anon.1990 & Anon.1999), here a more detailed investigation was done with the help of T.L.S, R.L.S and powder microscopy

Materials and methods

The plant material has been collected from Vadodara, Gujarat. Phytochemical analysis of roots of the plant for their secondary metabolites such as alkaloids, flavonoids, phenolics, steroids, quinones, mucilages and other compounds were done by standard methods of chromatography and spectrophotometry described in Chapter 2. Pharmacognostic, HPTLC finger printing and physico-chemical studies were done by using standard methods described in Chapter 2.

Results

Phytochemistry

Along with reported glycyrrhizin the root also showed the presence of good no.of phenolic acids such as vanillic, syringic, ferulic (*cis*- and *trans*-isomers) and *p*-coumaric acids. Mucilage amounted to 8.6 % consisting of galactose. The root also showed the presence of unidentified alkaloids and steroids.

Pharmacognosy

Macroscopic characters

The root was cylindrical and woody. The outer surface was yellowish brown to dark brown in colour and showed longitudinal wrinkles. Fracture fibrous , odour distinc, taste slightly acrid and sweet.

Microscopic characters

Root : T.S (Fig.83)

Cork, the outermost tissue, composed of 3 to 6 rows of thin walled, rectangular tangentially elongated cells with reddish brown contents of which the outermost one or two rows of cells were slightly ruptured and light brown in colour.A phellogen composed of a single row of narrow, thin walled, tangentially elongated cells. The phelloderm was not differentiated clearly. Cortex was made of narrow zone of thin walled parenchymatous cells. The cells were round to oblong in shape with some of having single monoclinic prisms (prismatic crystals) of calcium oxalate in

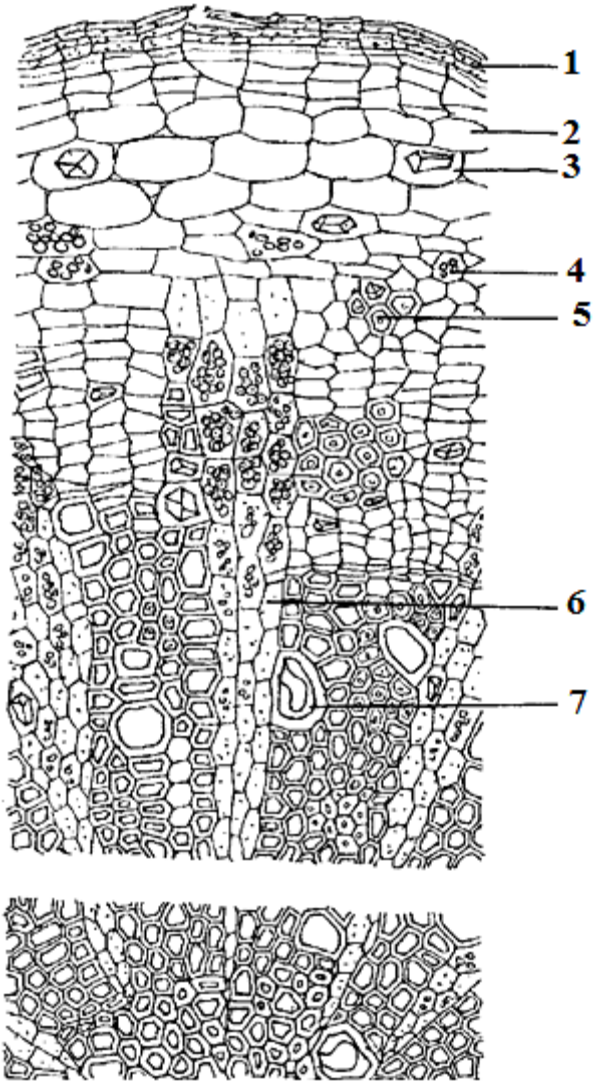


Fig.83.*Glycyrrhiza glabra* root, T.S: 1. Cork, 2. Cortex, 3. Parenchyma with prismatic crystals, 4. Starch grains, 5. Bast fibres, 6. Xylem rays,7. Vessels.

each cell. The bast was composed of an outer region wherein patches of bast fibres alternated with the regular phloem elements. Each patch of bast fibre contained up to abought 50 fibres. The bands of medullary rays present here is quite broad formed wedge shaped. The cambium was a distinct narrow band. The wide zone of wood composed mostly of secondary xylem. The wood consisted of vessels and patches of fibre tracheids alternating with patches of libriform fibres. Vessels showed presence of tyloses. The rays were filled with starch grains and prismatic crystal occasionally.

Root : T.L.S (Fig.84)

Cork cells appeared rectangular with brown contents. Large polygonal parenchyma contained prismatic crystal. The bast fibres were with a medium sized lumen. The rays were spindle shaped wherein the cell walls were pitted and filled with starch grains. The tracheids were with 2-3 rows of bordered pits. Vessels were many in number, having straight end walls showing bordered pits and reticulated thickened. In certain vessels, bordered pits were in compact rows and the pits were angular.

Root : R.L.S (Fig.85)

The phloem rays were upright and rectangular in shape and each cell was filled with starch grains. Here the bast fibres were found associated with prismatic crystals. Vessels showed the presence of tyloses.

Root : Powder study (Fig.86)

The components present in the powder were of cork of two types, parenchyma containing prismatic crystals and starch grains, crystals fibres, phloem fibres, vessels with angular bordered pits and reticulate thickened.

The rhizome of *G. glabra* which also used in medicine is similar to the root in almost all aspects,but differs in having a moderstely large pith in the centre.

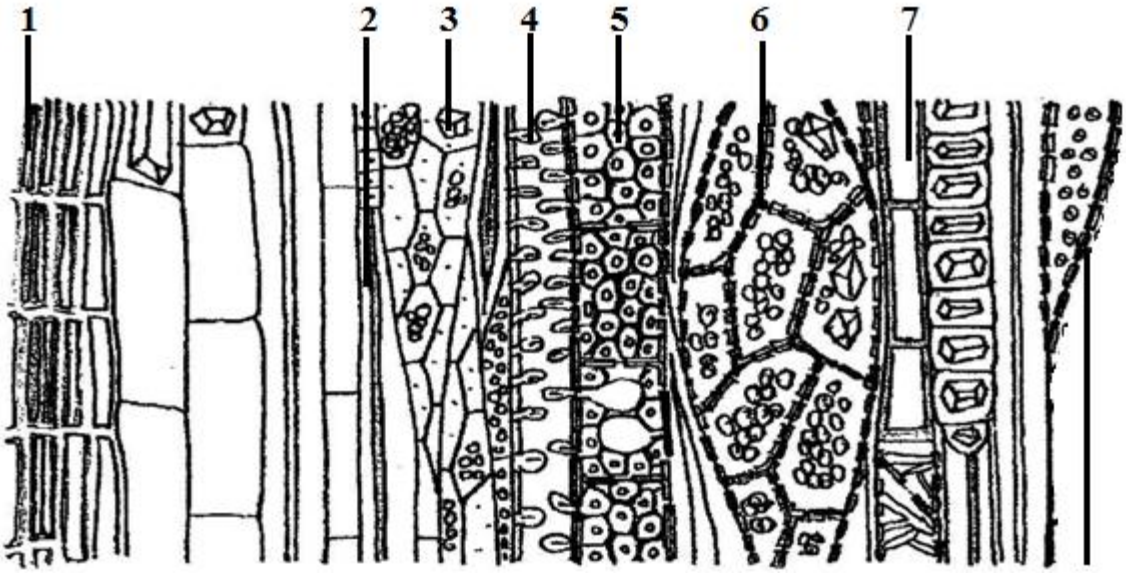


Fig.84.*Glycyrrhiza glabra* root, T.L.S:1.Cork, 2.Phloem fibres,3.Phloem rays,4. Trachieds, 5. Vessels with tyloses, 6. Xylem rays.7.Wood parenchyma.

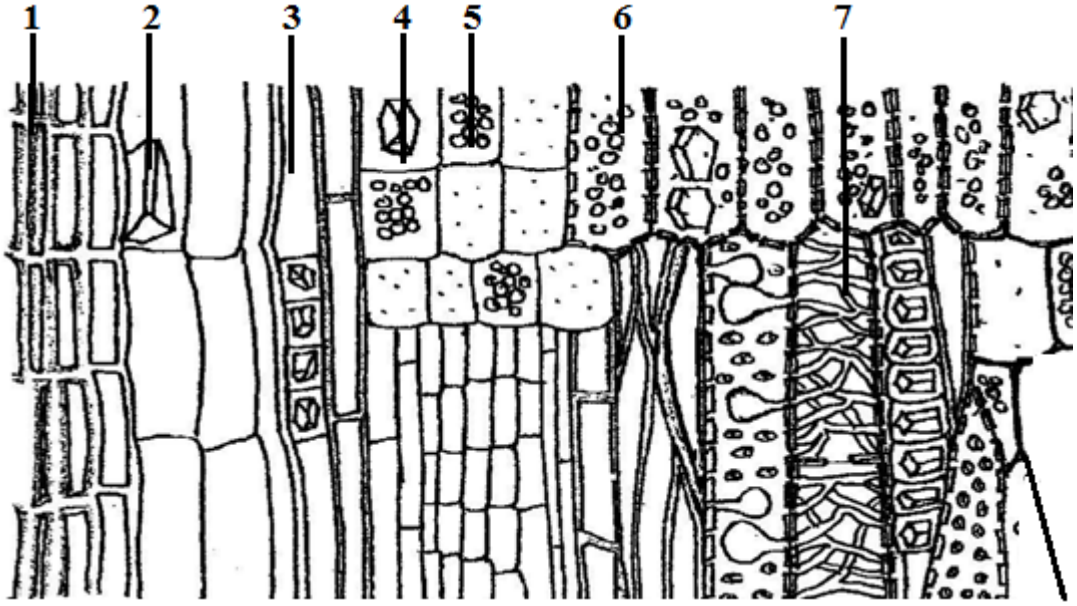


Fig.85.*Glycyrrhiza glabra* root, R.L.S:1.Cork, 2.Prismatic crystals, 3.Crystal fibres,4.Phloem rays with prismatic crystals, 5.Phloem rays with starch grains, 6.Xylem rays, 7. Reticulate thickened vessels.

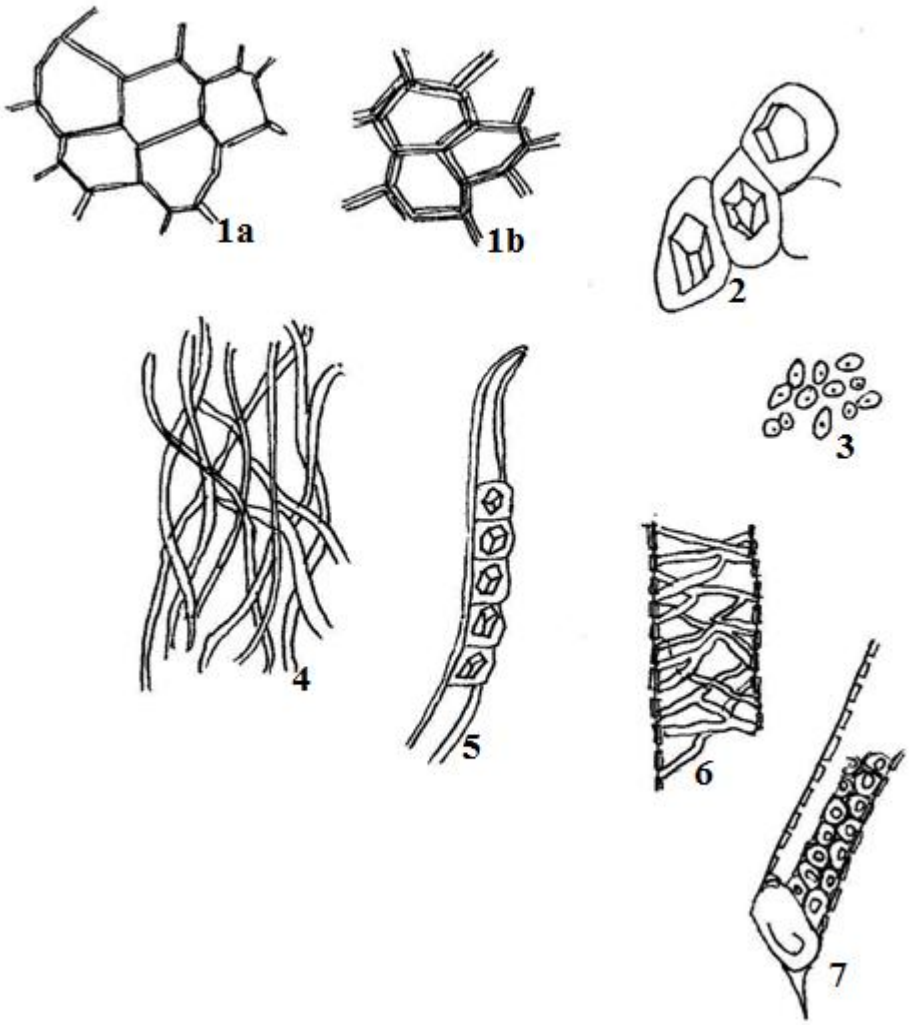


Fig.86.*Glycyrrhiza glabra* root, Powder study: 1.Cork a) Thin walled b) Thick walled, 2. Prismatic crystals, 3. Starch grains, 4. Phloem fibres, 5. Crystal fibres, 6. Reticulate thickened vessels. 7. Angular bordered pits in vessels.

Distinguishing features

Phytochemical markers

1. Ferulic (*cis*- and *trans*-isomers) acid.
2. *p*-Coumaric acid.

Pharmacognostic markers

1. Two types of cork cells i) thin walled and ii) thick walled.
2. Starch grains
3. Prismatic crystals.
4. Crystal fibres.
5. Vessels bordered pitted and reticulate thickened
6. Angular bordered pits in vessels
7. Presence of tyloses.

Physico-chemical analysis:

Table :14. Values obtained for the proximate analysis.

Sr.No.	Parameter	Mean ± SD (%)*			Average (%)
		Summer	Monsoon	Winter	
1.	Total ash content	06.31±0.19	06.26±0.20	06.29±0.16	6.29
2.	Acid insoluble ash content	01.69±0.21	1.73±0.33	01.69±0.26	1.70
3.	Alcohol soluble extractive	20.36±0.16	20.48±0.11	20.42±0.14	20.42
4.	Water soluble extractive	24.21±0.33	24.30±0.34	20.31±0.31	22.94

*Each value is a mean of 3 readings.

5.b.*Abrus precatorius.* Linn. (Fabaceae)

Sanskrit : Angaravalli, Aruna, Bhilabhushana, Gunja, Gunjika, Kaka, Kakachinchi.

Vernacular names:

Assamese : Latuwani , Aainuddik, Ratti Surkh.

Bengali : Kunch, Kawet.

English : Indian Liquorice,Coral Pea, Crab S Eye, Lucky Or Paternoster Bean.

Gujrati : Chanoti, Rati.

Hindi : Gunj, Chirmiti, Gaungchi, Gunja, Kunch, Rati, Tatti.

Kannada : Galaganji, Haga.

Malayalam: Atimadhuram,Cekkunni,Irattimadhuram,Kakani,Klitakam,Kunni.

Manipuri : Chaning.

Marathi : Gunj, Khaksi, Gunja.

Oriya : Runjo.

Persian : Chashmkhuros, Chashmekharush, Chashmkuros, Chashme-Khuros, Surkh.

Tamil : Gundumani, dimaduram, Adingam, Adisamiyai, Uyar, Uyarvukkoti.

Telugu : Atimadhuramu, Gurija, Gurivenda, Guruginja, Kukkutamu, Raktika, Mukkutamu.

Tibetan : Ma Ru Rtse, O La Mase Dmarpo.

Urdu : Ghunchi, Tukhm Kunch, Maghz Tukhm Kunch, Ghunchchi.

Distribution and habitat

This is a common plant occurring wild, found throughout tropical India and other warm countries from sea level up to 3000 feet under mesophytic conditions; seldom cultivated.

Morphological features.

A deciduous, wiry climber with tough branches: leaves abruptly pinnate with many pairs of leaflets, the rachis ending in a spine; the leaflets oblong, rounded at both ends, thinly membranous; flowers pink, clustered on tubercles arranged along the rachis of one-sided pedunculate raceme; fruits with a sharp deflexed beak; seeds usually scarlet with a black spot or sometimes pure white.

Medicinal uses:

The roots and leaves are astringent, sweet, emetic, diuretic and alexeteric. They are useful in cough, pharyngodynia, pectoralgia, inflammation, strangury and in vitiated conditions of vata.(Vaidyaratnam *et al.*,1994).

Previous Phytochemical reports

The roots contain glycyrrhizin, the active principle of liquorice(Anon.1948), abrasine,abrol (Khaleqe *et al.*,1966), choline, hypaphorine, precatorine(Ghosal *et al.*,1971),abrine.(Ghosal *et al.*,1971 & Karawaya *et al.*,1980), abruquinones(Kuo *et al.*,1995 & Kuo *et al.*,1999), abrusgenic acid, methyl abrusgenate, abruslactone A(Chiang *et al.*,1983), 7,5-dihydroxy-6,4-dimethoxy isoflavone and 7-*O*-*β*-D-galactopyranoside(Saxena *et al.*,1999).

Previous pharmacognostic reports

Only the T.S of the root has been done (Anon.1999 & Gupta *et al.*,2008) but study of T.L.S and R.L.S is remaining to be done. So a detailed study is conducted on root of the plant.

Materials and methods

The plant material has been collected from Pavagadh, Vadodara, Gujarat. Phytochemical analysis of roots of the plant for their secondary metabolites such as alkaloids, flavonoids, phenolics, steroids, quinones, mucilages and other compounds were done by standard methods of chromatography and spectrophotometry described in Chapter 2. Pharmacognostic, HPTLC finger printing and physico-chemical studies were done by using standard methods described in Chapter 2.

Results

Phytochemistry

There was no flavonoid in the root.The phenolic acids located were vanillic, syringic, ferulic (*cis*- and *trans*-isomers), melilotic, *p*- coumaric and *o*-coumaric acids. Mucilage amounted to 7.3% consisting of galactose. The root also showed the presence of unidentified alkaloids and steroids while coumarins and saponins were found in good concentrations.

Pharmacognosy

Macroscopic characters (Fig. 87)

The roots were vertical with few lateral roots, woody, cylindrical somewhat tortuous with minutely lenticellate warty surface and light brown in colour; odour not very particular. Taste faintly sweet. Fracture outer splintery, inner somewhat fibrous.

Fig.87. *Abrus precatorius* root.

Microscopic characters

Root : T.S (Fig. 88)

The outermost layer of the cork (28 to 48µ) consisted of 4 to 9 rows of rectangular to slightly tangentially elongated cells with yellowish-brown contents. The cells were tightly fitted together one above the other and without intercellular spaces. The outer layers of the cork were broken at places. The phellogen was indistinct. The cortex was a thin zone consisting of two to four rows of tangentially elongated large cells, some of which contained starch grains and a few prismatic crystals and was characterized by the presence of prominent, ring of sclereids, composed of oblong, ovoid or radially elongate stone cells with thick pitted walls. Some of them contained starch grains. Adhering to both the inner and outer margins of this ring and spaced at short intervals were parenchyma cells found containing prismatic crystals. The bast made up of usual elements and the cells of outer layers were comparatively large in size then that of inner layers. Here there were groups of bast fibres composed mostly of 2 to 4 or more tangential bands of thick walled

(double layered) gelatinous fibre groups, alternating with the regular phloem elements and being intercepted radially by the medullary rays. The wood was composed of many vessels, xylem parenchyma, fibre tracheids and the medullary rays. The wood fibres found in patches alternating with parenchyma, The vessels occurred mostly in pairs. Medullary rays were bi-seriate to multi-seriate (upto 12 rows) and most of the cells were packed with starch grains while a few other contained prismatic crystals of calcium oxalate. The ray cells were fairly thick walled and having simple pits on their walls and were usually tangentially elongated.

Root : T.L.S (Fig. 89)

The bark cells were seen squarish to rectangular in shape, some of which were filled with brown content. Cortical parenchyma were hexagonal in shape contained prismatic crystal and starch grains. The bast fibres had a very thin distinct lumen. The phloem rays were filled with starch grains. Xylem rays were mostly broad multiseriate, spindle shaped wherein the cell walls were thick, pitted and fully filled with starch grains and isolated prismatic crystal. Some of the cells in the center were found to be curved.

Root : R.L.S (Fig. 90)

The cork cells were few layered. Below cork in the cortex were radial rows of stone cells. The phloem cells contained starch grains and prismatic crystals. The bast fibres appeared straight in R.L.S. the xylem rays were pitted and contained starch grains inside the cells. The xylem tracheids were bordered pitted.

Root : Powder study (Fig. 91)

The components present in the powder were cork cells, fragments of cortical parenchyma, starch grains, prismatic crystals, stone cells, crystal fibres, ray parenchyma, fibre tracheids and bordered pitted vessels.

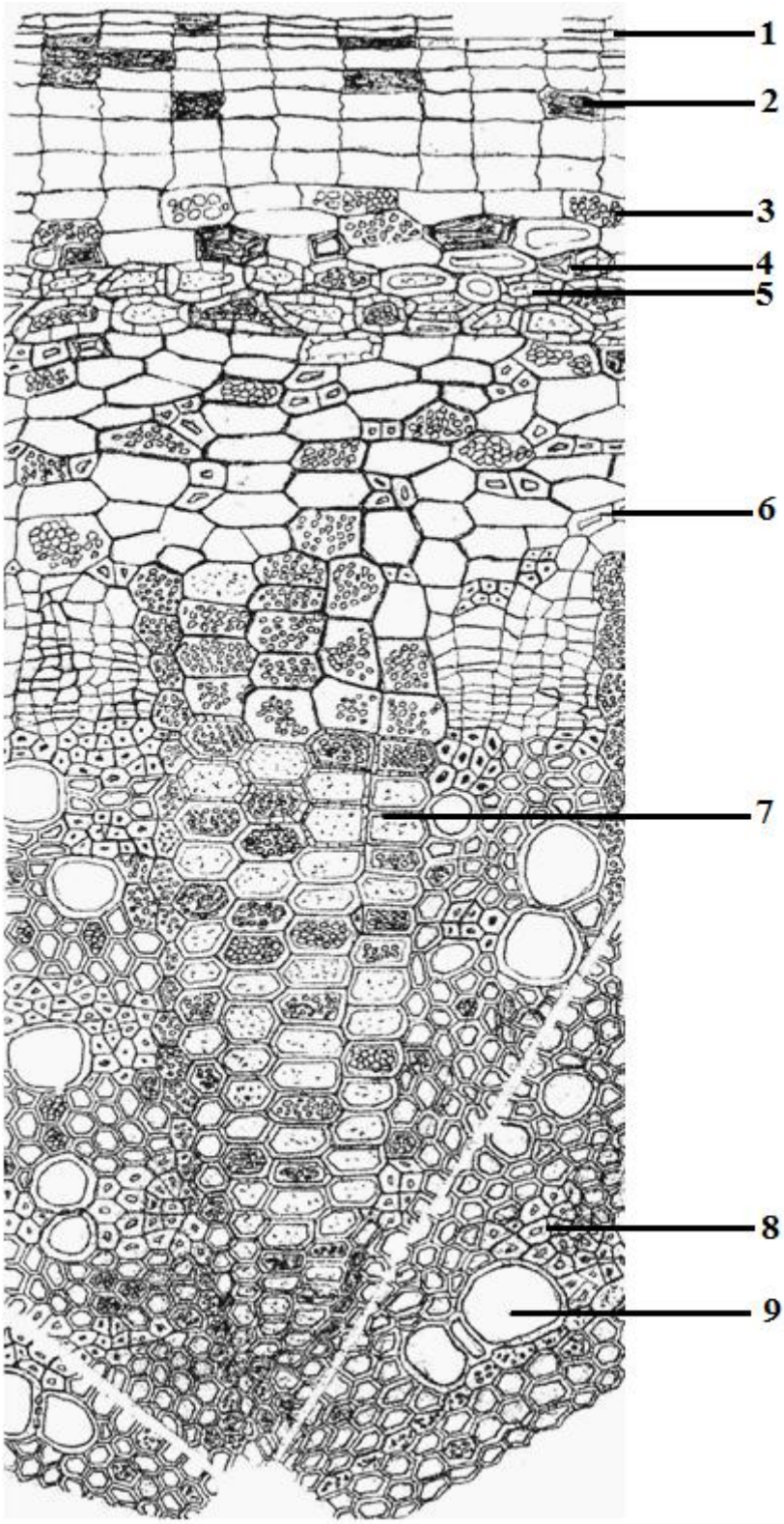


Fig.88.*Abrus precatorius* root, T.S: 1. Cork, 2. Cork with yellowish-brown contents 3. Parenchyma with starch grains,4. Prismatic crystals, 5. Stone cells, 6. Bast fiber,7.Xylem rays, 8.Libriform fibres, 9. Vessels.

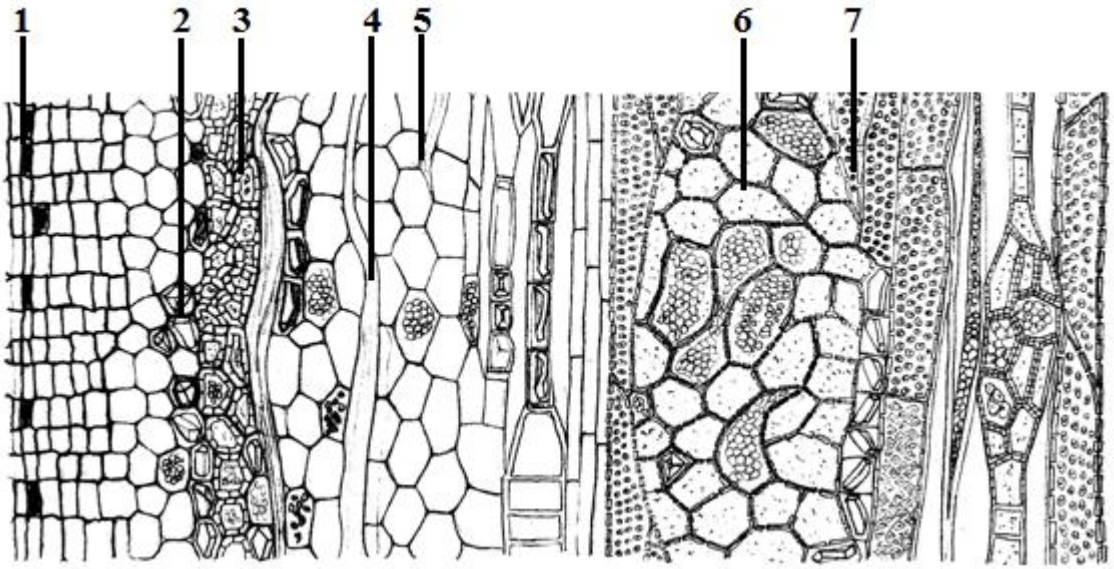


Fig.89. ***Abrus precatorius*** **root, T.L.S:** 1. Cork cells, 2. Prismatic crystals, 3. Stone cells,4. Bast fibres, 5. Phloem rays, 6. Xylem rays, 7.Fibre tracheids.

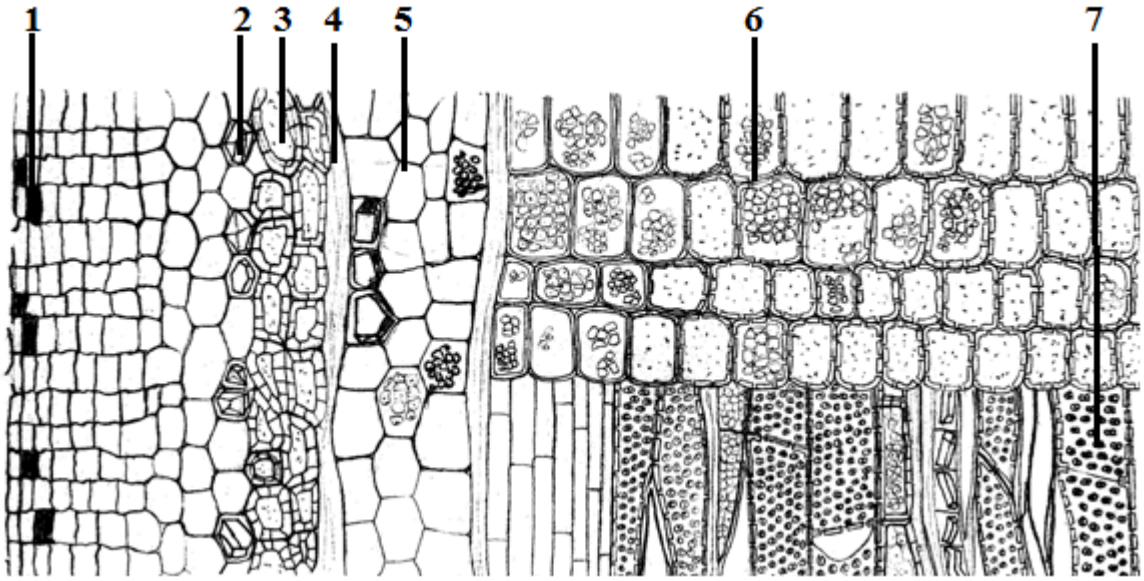


Fig.90. ***Abrus precatorius.*** **Linn root, R.L.S:** 1. Cork cells, 2. Sphaeraphides, 3. Stone cells,4. Bast fibres, 5. Phloem rays with starch grains and prismatic crystals, 6. Xylem rays, 7.Vessels.

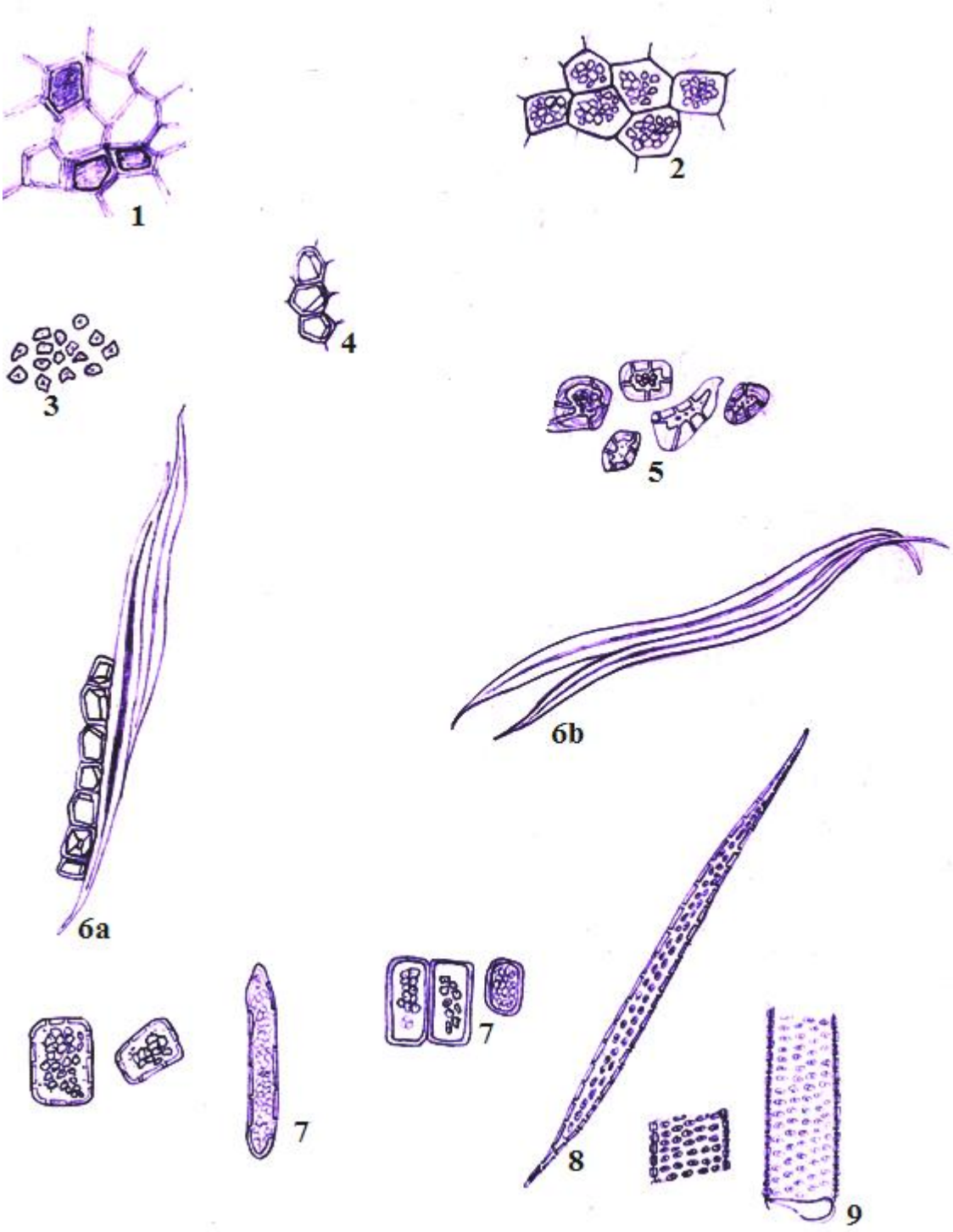


Fig.91. *Abrus precatorius.* Linn root, Powder study: 1. Cork cells, 2.Fragments of cortical parenchyma, 3.Starch grains, 4.Prismatic crystals, 5.Stone cells, 6.a)Crystal fibres, b)Wood fibres, 7.Ray parenchyma with starch grains, 8.Fibre tracheids, 9. Bordered pitted vessels.

Distinguishing features

Pharmacognostic markers

1. Starch grains.
2. Prismatic crystals.
3. Stone cells.
4. Crystal fibres.
5. Cells of ray parenchyma in the center were found to be curved in T.L.S.

Phytochemical markers

1. Ferulic (*cis*- and *trans*-isomers) acid.
2. Melilotic acid.

Physico-chemical analysis:

Table 15: Values obtained for the proximate analysis.

Sr.No.	Parameter	Mean ± SD (%)*			Average (%)
		Summer	Monsoon	Winter	
1.	Total ash content	04.73±0.16	04.81±0.21	04.49±0.26	4.68
2.	Acid insoluble ash content	01.07±0.25	0.99±0.54	01.08±0.29	1.05
3.	Alcohol soluble extractive	14.03±0.16	14.33±0.21	14.12±0.18	14.16
4.	Water soluble extractive	10.23±0.33	10.01±0.22	10.18±0.36	10.14

*Each value is a mean of 3 readings

5.c. *Alysicarpus longifolius* (Rottl. Ex Spreng.) Wight & Arn. (Fabaceae)

Vernacular names:

Gujrati : Ghoda samerwo, Ubhosamerwo, Dhodasamervo.

Hindi : Jangali gailia, Gubal.

Marathi : Shevra, Motha dampta.

Tamil. : Naamappoondu.

Telugu : Peddakandikaraku.

Distribution and habitat

It is distributed in Saurashtra, Madhya Pradesh, Bombay, Madras.

Morphological features

The plant is an erect herb 1.2-1.5 m tall. Leaves unifoliolate; stalks 3-10 mm long, leaflets 5-15 X 1-2 cm, oblong or lanceolate, base heart-shaped. Inflorescence is a dense raceme, 15-30 cm long. Bracts often longer than 1.3 cm, ovate, pointed. Flowers 1 cm, in pairs, blooming from the base of the spike upwards. "Standard" petal is yellow flushed with red. Wing and keel are dark pink. Pods 1 cm, 4-6 jointed.

Previous Phytochemical reports

The root is sweet and has been reported as a substitute for liquorice (Nadkarni, 1954; Chopra *et al.*,1956; Wealth of India. 1985).The ethanolic extract of the leaves and its various fractions were found to yield myricy1 alcohol, β-sitosterol, β-sitosterol acetate, rutin and pinitol (Jain and Gupta.1981). The flavonoid glycosides isolated were 4' –α-D-glucopyranosyloxy-5-hydroxy-7-methoxyflavone (Jain and Gupta, 1984).quercetin-7-*O*-rhamnoside, chrysoeriol-7-*O*-xyloside and kaempferol-3-*O*-xyloside-7-*O*-rhamnoside(Jain and Gupta. 1986b) .The seed was found to be rich in amino acids. These were glutamic acid, aspartic acid, arginine, leucice, lysine, serice, phenylalanine, proline, valine, glycine, isoleucine, alanine, tyrosine, threonine, histidine, cystine and methionine. The seed oil showed the presence of saturated and unsaturated fatty acids (Jain and Gupta, 1985).

Previous pharmacognostic reports

No study has been done on the pharmacognostic characters of root of the plant.

Materials and methods

The plant material has been collected from Timbi, Baroda, Gujarat. Phytochemical analysis of roots of the plant for their secondary metabolites such as alkaloids, flavonoids, phenolics, steroids, quinones, mucilages and other compounds were done by standard methods of chromatography and spectrophotometry described in chapter 2. Pharmacognostic, HPTLC finger printing and physico-chemical studies were done by using standard methods described in Chapter 2.

Results

Phytochemistry

The roots of the plant along with saponin, steroids were found to contain vanillic and syringic acids while melilotic acid and flavonoids were found to be present in traces. Mucilage amounted to 2.07 % consisting of rhamnose and xylose.

Pharmacognosy

Macroscopic characters (Fig.92)

The roots were vertical, woody, somewhat tortuous and of a pale buff colour; odour not very particular. Taste slightly bitter. Fracture short.

Fig.92. *Alysicarpus longifolius* root.

Microscopic characters

Root : T.S (Fig.93)

The T.S of the root was circular in outline with a large central woody region and a thin outer bark. The cork consisted of 2 to 4 rows of thin walled, tangentially

elongated cells. The phellogen was indistinct. The cortex was very narrow made up of three to six rows of comparatively large polygonal or tangentially elongated thin walled parenchyma cells. Some of them become thick walled collenchymatous. Isolated prismatic crystals and starch grains were found present in the cortex. Bast fibres were in a small patches, composed of two to ten or more thick walled (double layered) gelatinous fibres, alternating with thin walled phloem elements along with isolated narrow lumen fibers. The number of gelatinous fibres in patches were more towards cortex than that of patches present towards the wood. Phloem parenchyma cells at the outer region of phloem were bigger than that of inner ones some of them contained prismatic crystals. Cambium was indistinct. Wood consisted of vessels, tracheids, fibres and rays. They varied in size and shape. Medullary rays were radially elongated and contained starch grains and a single sphaeraphides in a cell. Here few isolated groups of gelatinous fibres were also found. Xylem in the centre were typically composed of thick walled fibre-tracheids. Vessels were many distributed throughout or occurring singly or many in a group of two. The association of pitted parenchyma with vessels were very common here. Starch grains were small, spherical or ovoid with a hilum in the centre.

.**Root : T.L.S (Fig.94)**

Cork cells appeared rectangular. The cells of the cortex were also appeared rectangular as of cork , but were larger in size. The phloem fibres were straight. Xylem rays were fairly thick walled, oval, simple pitted and each cell contained 10-15 starch grains and 1-3 prismatic crystals each. The vessels were found attached with radial rows of pitted parenchyma. Tracheids contained 2 to 3 rows of bordered pits. The bordered pits in vessels were arranged compactly in 5-6 rows. The primary xylem had both annular and spiral thickenings.

Root : R.L.S (Fig.95)

The phloem ray cells appeared rectangular. They were thin walled and filled with 10-15 starch grains and 1-2 prismatic crystals each cell. The xylem rays also were filled with starch grains and sphaeraphides. The pits on the wall were of simple type. The vessels showed 3-4 rows of bordered pits. The primary xylem showed spiral thickenings.

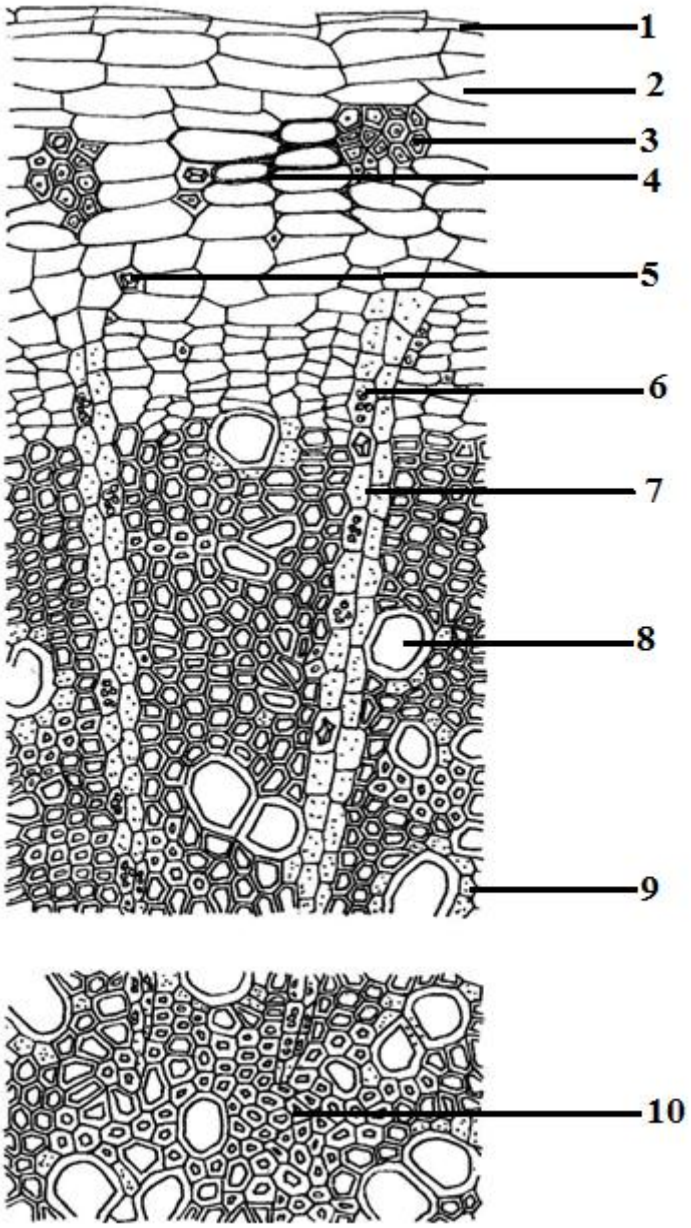


Fig.93.*Alysicarpus longifolius* root, T.S: 1. Cork, 2.Cortex, 3. Gelatinous fibres, 4. Collenchyma, 5.Prismatic crystals, 6.Starch grains, 7.Xylem rays, 8.Vessels, 9. Pitted parenchyma, 10. Fibre-tracheids.

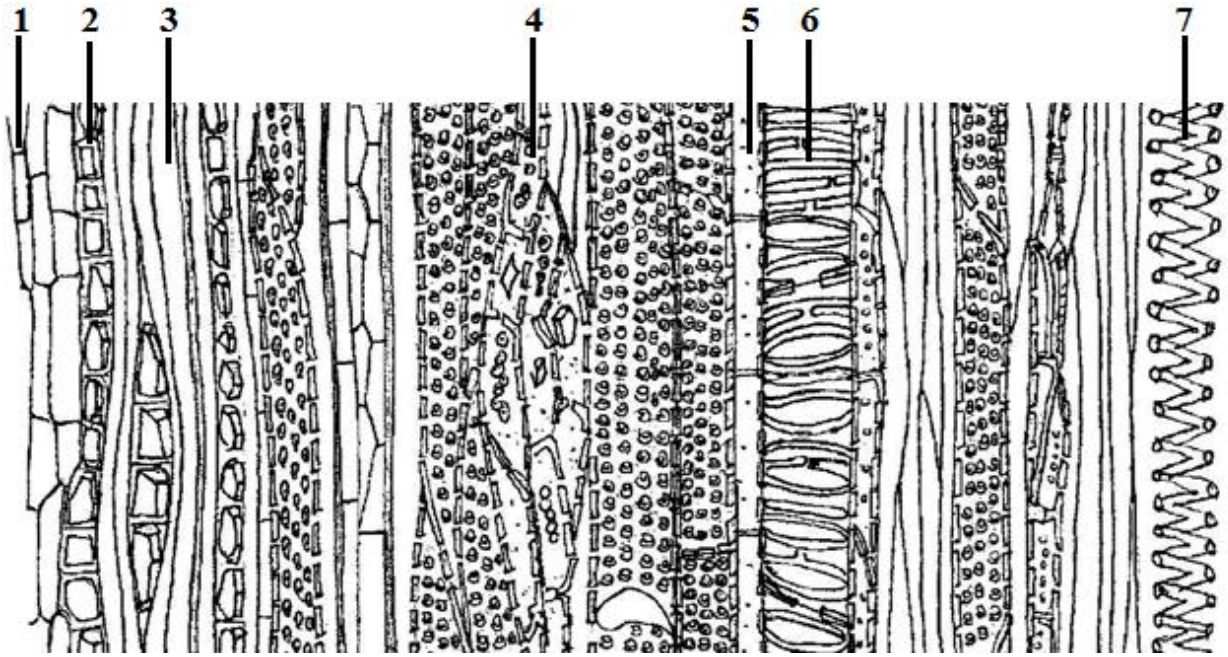


Fig.94. ***Alysicarpus longifolius*** **root, T.L.S:**1. Cork, 2.Prismatic crystal,3. Phloem fibres,4. Starch grains, 5. Pitted parenchyma, 6. Vessels with annular thickening, 7. Spiral thickened vessels.

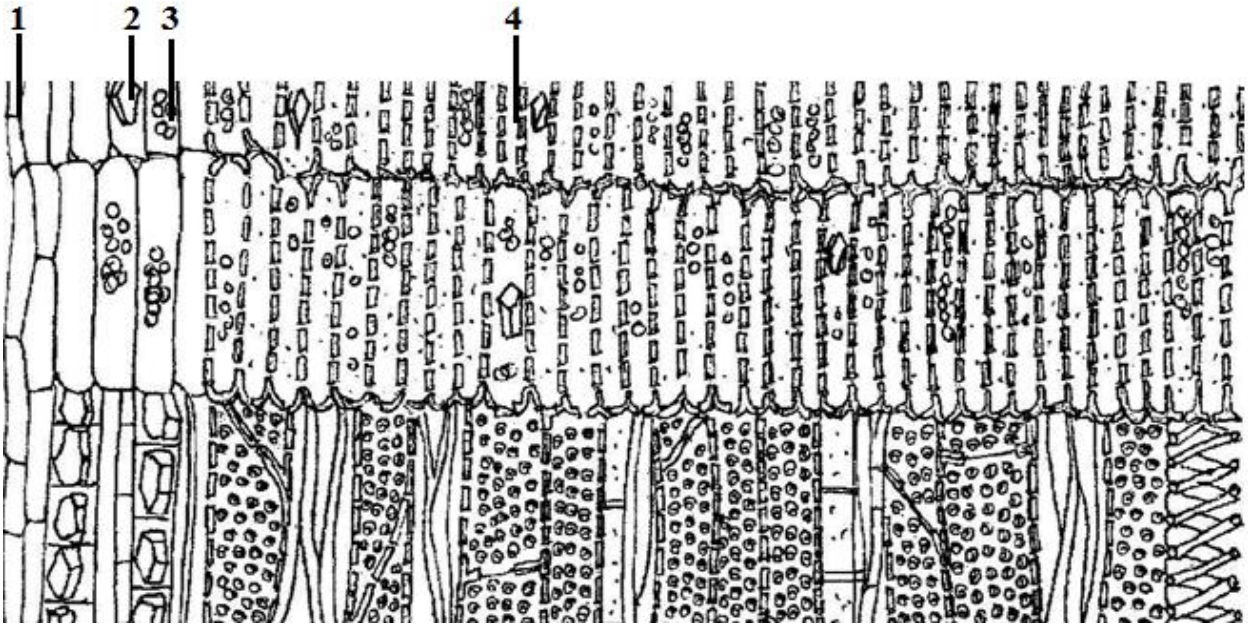


Fig.95. ***Alysicarpus longifolius*** **root, R.L.S:**1. Cork cells, 2.Prismatic crystal,3. Phloem rays with starch grains and prismatic crystals, 4. Xylem rays with starch grains and prismatic crystals.

Root : Powder study (Fig.96)

The components present in the powder were thin walled cork, prismatic crystal, starch grains, gelatinous fibres, crystals trapped between fibers, ray parenchyma with prismatic crystal , annular thickened tracheids.

Distinguishing features

Pharmacognostic markers

1. Thin walled cork.
2. Prismatic crystals.
3. Starch grains.
4. Gelatinous fibers.
5. Ray parenchyma containing prismatic crystal and starch grains.
6. Bordered pitted vessels.
7. Spiral and annular thickened tracheids.

Phytochemical markers

1. Vanillic acid.
2. Syringic acid.
3. Rhamnose.
4. Xylose.

Physico-chemical analysis:

Table 16 : Values obtained for the proximate analysis.

Sr.No.	Parameter	Mean ± SD (%)*			Average (%)
		Summer	Monsoon	Winter	
1.	Total Ash Content	02.42±0.22	02.63±0.32	02.43±0.38	2.49
2.	Acid Insoluble Ash content	0.98±0.31	1.07±0.66	1.01±0.39	1.02
3.	Alcohol soluble extractive	03.12±0.10	03.09±0.19	03.16±0.13	3.12
4.	Water soluble extractive	05.23±0.31	05.46±0.16	05.31±0.06	5.33

*Each value is a mean of 3 readings

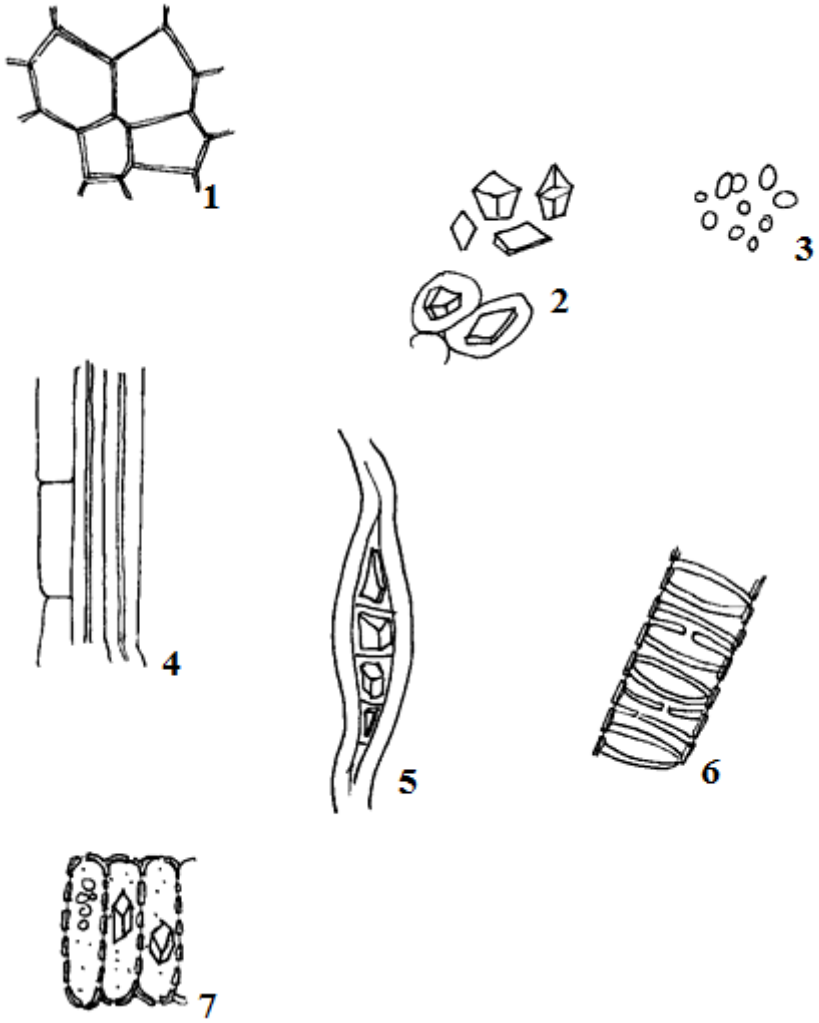


Fig.96. ***Alysicarpus longifolius*** **Powder study:** 1.Cork, 2.Prismatic crystal, 3.Starch grains, 4.Gelatinous fibres, 5.Crystals trapped between fibers, 6.Vessels with annular thickening,7.Xylem rays with starch grains and prismatic crystals.

5.d.*Maerua arenaria* Hook. f. &Thoms. (Capparidaceae)

Synonyms: ***Niebuhria arenaria* DC.Prodr.**

Vernacular names:

English : Earth Sugar-root.

Hindi : Vika.

Gujarati : Morinika, Dholokatkiyo, Dudhiyohemkand,Hemkand .

Tamil : Pumi Carkkarai Kilantu, Pumicarkkaraik.

Telugu : Bhumichakkarai Pattatiga, Bhucakramu.

Tibetan **:** Ro Ma Ha.

Distribution and habitat

The plant is a large woody climber, found in Southern and Central India, and Ceylon.

Morphological features.

The plant is climbing woody shrub with divaricate branches; bark smooth, pale. Leaves 2.5- 5 by 1-2.5 cm, elliptic-oblong, mucronate, glabrous. Flowers in corymbs, greenish-white, terminal or on lateral shoots. Bract one at the base of each pedicel, small, ovate. Calyx-lobes ovate, hooded at the apex, with a short horn behind the hood. Petals ovate-lanceolate, acute, with slightly undulate margins. Stamens many, inserted on the torus. Gynophore 2 cm long. Ovary cylindric, truncate; style 0; stigma large. Fruit pale brown, constricted between seeds. Seeds brown, globose, echinulate.

Medicinal uses:

The root is said to be used as an alternative, tonic, and stimulant.(Anon.1990) and for bleeding piles, as alterative in fevers; as a tonic in muscular debility.(The root resembles liquorice root in appearance and taste) (Khare,2007).

Previous Phytochemical reports

No detailed phytochemical studies have been done on root of this plant. The plant contained ordinary plant constituents and a certain quantity of sugar.(Nadkarni 1954).

Previous pharmacognostic reports

No study has been done on the pharmacognostic characters of root of the plant.

Materials and methods

The plant material has been collected from Rajpipala,Gujarat. Phytochemical analysis of roots of the plant for their secondary metabolites such as alkaloids, flavonoids, phenolics, steroids, quinones, mucilages and other compounds were done by standard methods of chromatography and spectrophotometry described in Chapter 2. Pharmacognostic, HPTLC finger printing and physico-chemical studies were done by using standard methods described in Chapter 2.

Results

Phytochemistry

The root was found to contained vanillic, syringic and melilotic acids. Mucilage amounted to 7.3% consisting of xylose. The root also showed the presence of unidentified alkaloids and steroids while coumarins and saponins were found in good concentrations. Saponins, Steroids and alkaloids were also present.

Pharmacognosy

Macroscopic characters (Fig.97)

The roots were vertical, somewhat tortuous and of a pale yellow in colour; odor not very particular, Taste slightly sweet. Fracture short.

Fig.97. *Maerua arenaria* root

Root : T.S (Fig.98)

The root possessed a few (4-9) layered cork where the cells were almost squarish .Outer 2-3 layers of phelloderm was of thick walled cells and the walls were

reddish-brown in colour. The cells of the phellogen was thin walled. Below the cork there was a single discontinues ring of a stone cells. The stone cells were small, circular to oblong, thick walled narrow lumened. Cortex made up of compactly arranged parenchymatous cell. The cells were polygonal in shape and showed the deposition of oil globules in it. The single or a group of stone cells were also embedded in the cortex. The stone cell were thick walled with distinct striations and narrow lumened filled with yellowish brown contents. There was single vascular bundles were also scattered in the cortex. Pericycle was made up of three to five layers of discontinuous rings of thick sclerenchyma. Phloem was 5-10 layered made up of usual elements with some of the cells showed the deposition of oil globules. The medullary rays were uni to biseriate and the cells were pitted some of contained oil globules. The xylem made up of tracheids with less no of wood parenchyma. The Vessels were many, scattered and were bordered pitted.

Root : T.L.S (Fig. 99)

Cork cells appeared rectangular with reddish-brown thick wall. The cells of the cortex were big polygonal and contained oil globules. The stone cells were rectangular and found filled with yellowish- brown content. Pericyclic sclerenchyma were straight. Phloem ray were thin walled and pitted. Fibre tracheids were pitted, and contained simple as well as bordered pits. Xylem rays were spindle shaped. The vessels were broad, with 3-4 alternate rows of bordered pits.

Root : R.L.S (Fig.100)

Cork cells were rectangular with reddish-brown thick wall. The phloem ray cells were erect, polygonal and were filled with oil globules. There were 3 to 4 companion cells found attached with each phloem parenchyma. Xylem rays were hexagonal, pitted and showed deposition of oil globules.

Root : Powder study (Fig.101)

The components present in the powder were thick walled cork cells, group of stone cells fragments of cortical parenchyma filled with oil globules, phloem ray cells with deposits, sieve tubes with deposits, bordered pitted vessels.

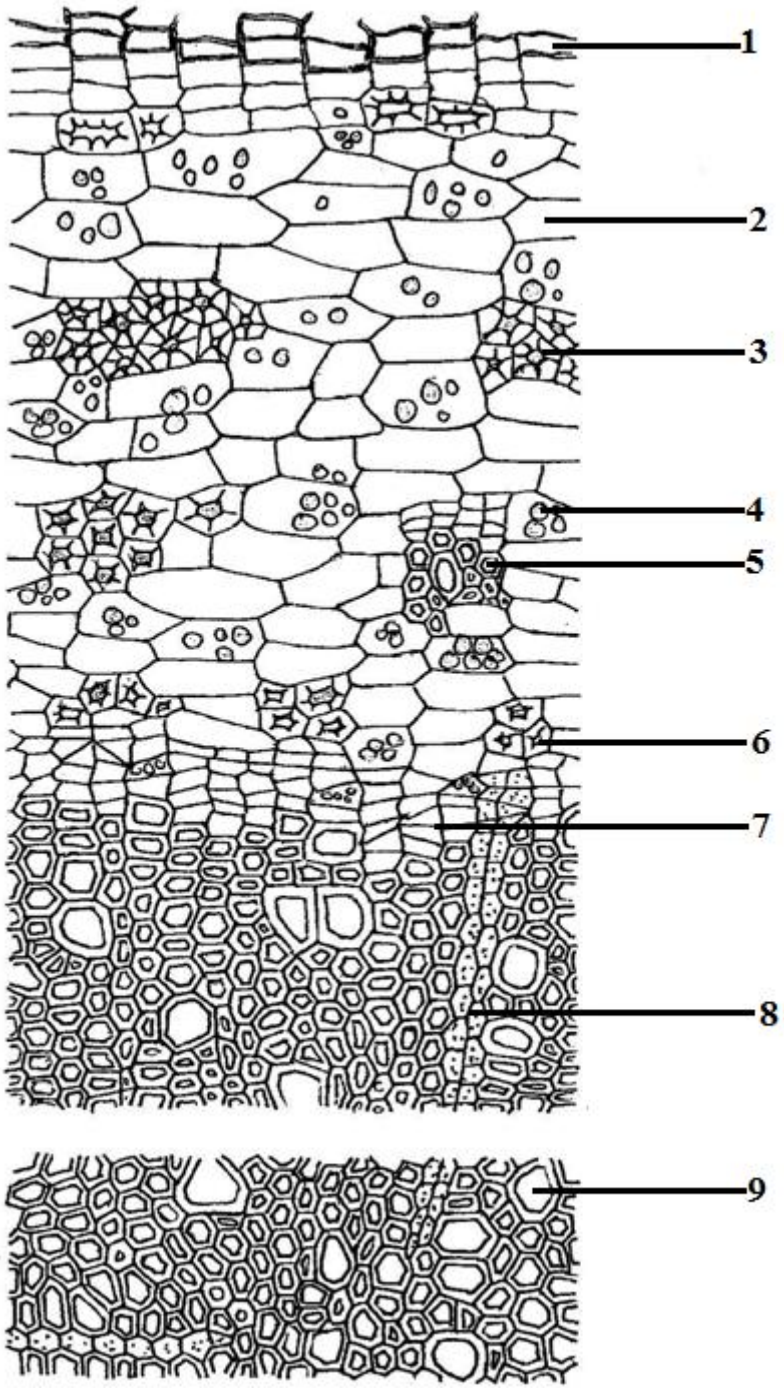


Fig.98 . ***Maerua arenaria*** **root, T.S:** 1. Cork, 2. Cortex , 3. Stone cell, 4. Oil globules ,5. Vascular bundle, 6. Pericycle,7. Phloem, 8. Xylem rays, 9. Vessels.

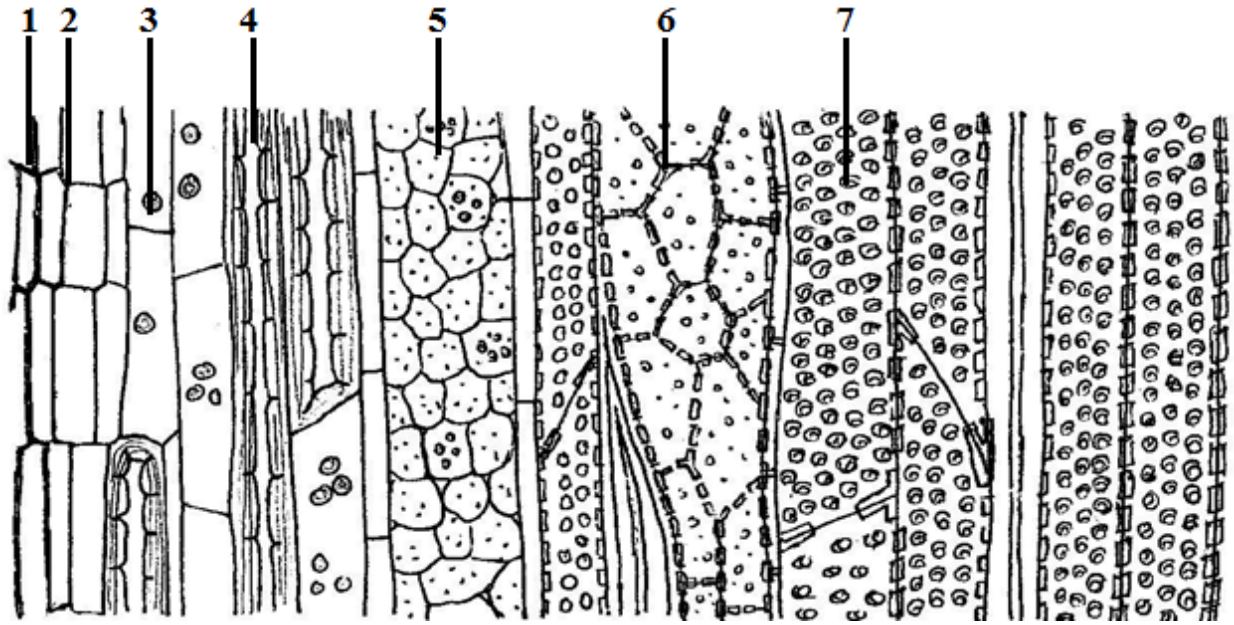


Fig.99. ***Maerua arenaria*** **root, T.L.S**:1.Cork cells with thick walls, 2.Cortex, 3.Oil globules,4. Stone cell, 5.Phloem ray, 6.Xylem rays, 7.Vessels with alternate bordered pits.

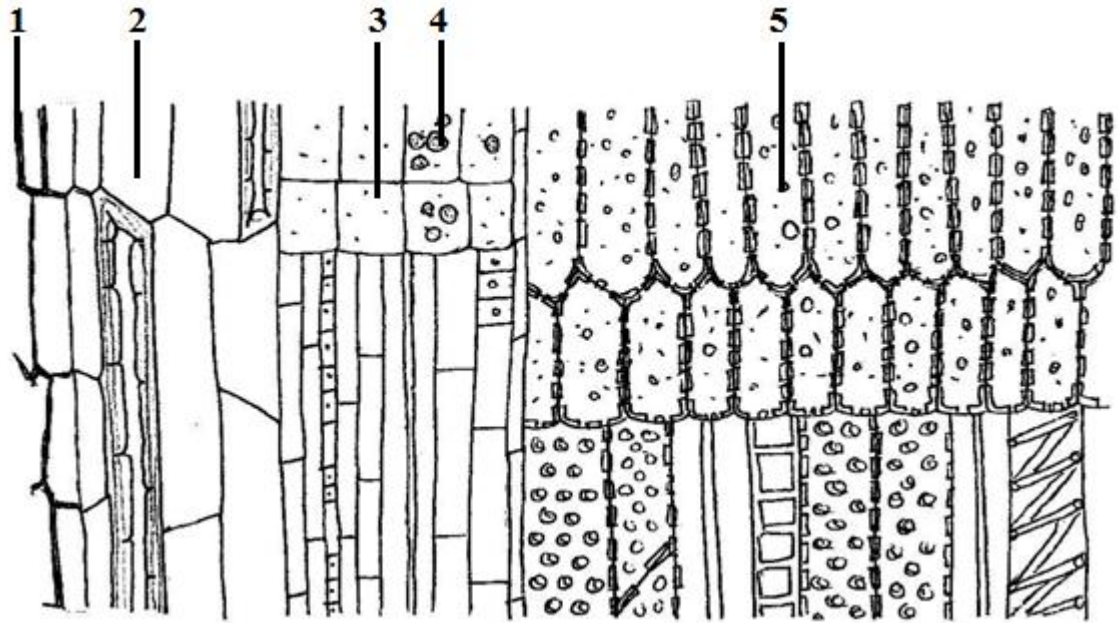


Fig.100. ***Maerua arenaria*** **root, R.L.S**:1.Cork cells, 2.Cortex, 3. Phloem rays, 4.Oil globules, 5. Xylem rays.

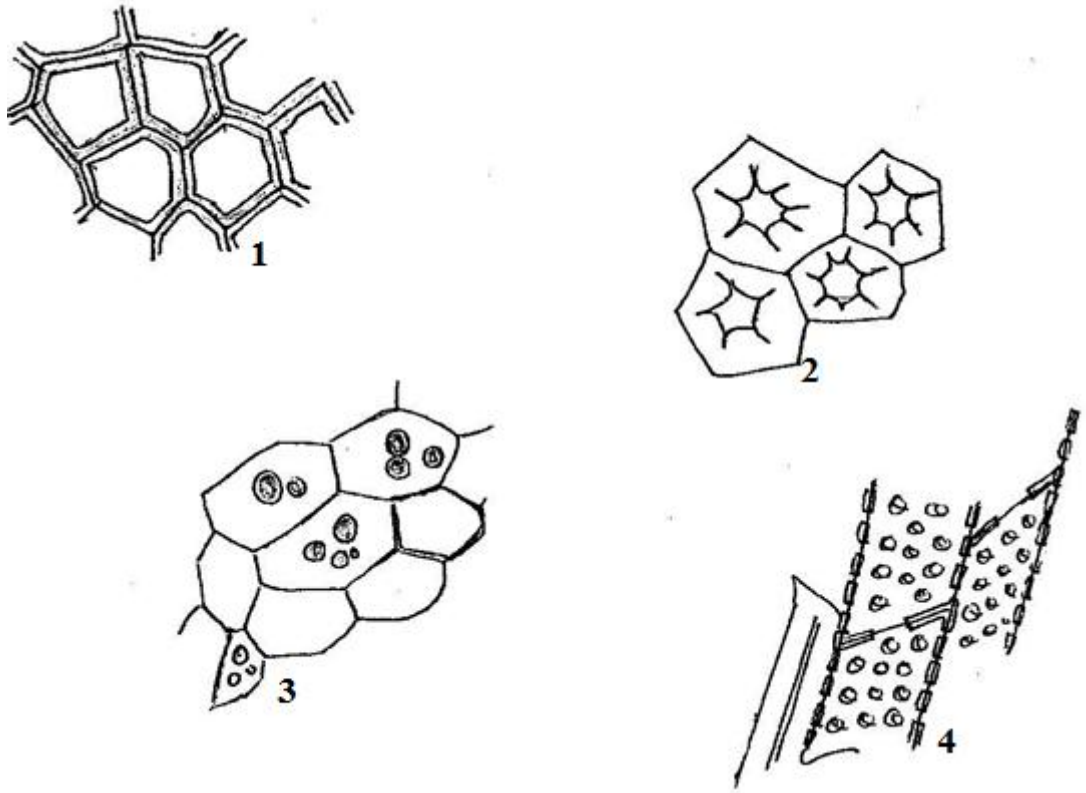


Fig.101 . ***Maerua arenaria*** **root, Powder study:** 1. Thick walled cork cells, 2.Stone cells, 3. Parenchyma filled with oil globules, 4. Bordered pitted vessel.

Distinguishing features

Pharmacognostic markers

1. Thick walled cork cells.
2. Parenchyma filled with oil globules.
3. Stone cells.
4. Bordered pitted vessel.

Phytochemical markers

1. Melilotic acid.

Physico-chemical analysis:

Table 17 : Values obtained for the proximate analysis.

Sr.No.	Parameter	Mean ± SD (%)*			Average (%)
		Summer	Monsoon	Winter	
1.	Total ash content	02.11±0.16	02.18±0.12	02.09±0.21	2.13
2.	Acid insoluble ash content	00.60±0.08	0.63±0.09	00.59±0.06	0.61
3.	Alcohol soluble extractive	10.01±0.21	10.13±0.23	10.02±0.13	10.05
4.	Water soluble extractive	08.19±0.42	08.16±0.02	08.24±0.29	8.2

*Each value is a mean of 3 readings**.**

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5.e.*Taverniera cuneifolia* (Roth) Arn. (Family – Fabaceae)

Synonyms: ***Taverniera abyssinica*** A.Rich, ***T. nummularia*** Baker non-DC.

Vernacular names:

English : East Indian Moneywort.

Gujrati : Jangali Jethimadh.

Hindi : Jangali Jethimadh.

Marathi : Jethi-madh.

Distribution and habitat

Plains of Punjab, Gujarat and the Deccan in waste places. (Khare,2007)

Morphological features

The plant is shrub, 60-100 cm, branches pubescent. Leaf uni-trifoliolate, leaflets 0.6-2.5 cm long, obovate to oblanceolate, entire, mucronate, pubescent, becoming subglabrous; stipules connate, amplexicaul, c. 3 mm long. Inflorescence an axillary raceme, up to 10 cm long, Pedicel 1-2.5 mm long, bracts c. 2.5 mm. Calyx 4-5 mm long, silky, teeth deltoid, c. 2.5 mm long. Corolla purple, macrescent. Vexillum 10-13 mm long, vexillum and keel larger than the wing. Fruit with 1-3, 1-seeded joints, joints echinate and ovoid, pubescent.

Medicinal uses

Leaves of this plant used as a poultice for sloughing wounds. Roots used as a substitute for liquorice. (Khare,2007) and exhibited promising anti-inflammatory, anti-tumor, anti germ tube formation (in *Candida albicans*),protection from mutagen toxicity and cytotoxic activities (Zore 2008).

Previous Phytochemical reports

The roots contained 13.20% glycyrrhizin.(Zore 2008)

Previous pharmacognostic reports

No complete study has been done on the pharmacognostic characters of root of the plant.

Materials and methods

The plant material has been collected from Surendranagar road,Gujarat. Phytochemical analysis of roots of the plant for their secondary metabolites such as alkaloids, flavonoids, phenolics, steroids, quinones, mucilages and other compounds were done by standard methods of chromatography and spectrophotometry described

in chapter 2. Pharmacognostic, HPTLC finger printing and physico-chemical studies were done by using standard methods described in Chapter 2.

Results

Phytochemistry

The phytochemical analysis of root of plant showed that it is rich in containing phenolic acids such as vanillic, syringic, ferulic, *o*-coumaric, melilotic , and *p*-hydroxy benzoic acids. It also showed the presence of coumarins,saponins,steroids and contained alkaloids and flavonoids in traces, while its mucilage contained sugar acid.

Pharmacognosy

Macroscopic characters (Fig.102):-

The roots occured in long, cylindrical slender pieces. The young root were light yellow, little shiny and comparatively smooth non-lenticellate surfaced with faint longitudinal fissures whereas the thicker mature pieces surface was grayish brown or light brown coloured and rough due to longitudinal fissures and transversely elongated wrinkles. The fracture was fibrous externally and hard in the centre. The roots were faintly sweet.

Fig.102. *Taverniera cuneifolia* root.

Microscopic characters

Root : T.S (Fig.103)

The root in transverse section was almost circular in outline with slightly undulating margin. The cork was composed of 3-6 rows of rectangular, tangentially

elongate cells. Those of the outer rows were usually much compressed and have thick light brown walls but the inner cells were arranged in regular rows and had comparatively thin light pinkish-brown walls and appeared small rectangular. The phellogen cells were found to be in collapsed condition. The phelloderm, consisted of usually 1 to 3 layers of thin walled parenchyma. The secondary cortex made up of 4 to 9 layers of thin walled tangentially elongated polygonal parenchymatous cells arranged compactly and lack in intercellular spaces, few of these cells were typical found in a pairs wherein the adjoining walls were straight and also showed the presence of small isolated groups of fibres of about 2 to 4, associated with cells containing isolated prismatic crystal of calcium oxalate. Most of the cells except a few rows towards the outside were filled with the starch grains. Cambium was indistinct. The secondary phloem was 9-12 layered. Besides having usual phloem elements it showed the presence of distinct groups of bast fibers arranged radially in groups of about 10 to 50 fibres, adhering to which are cells of crystal fibres containing prisms of calcium oxalate and gelatinous fibre. The phloem parenchyma present towards the cortex was comparatively larger than that of inner one present towards the wood. Phloem rays were thick walled pitted parenchymatous and the cells were tangentially elongated rectangular to polygonal in outline and slightly larger than the phloem parenchyma cells, loaded with starch grains. Xylem was dominated by fibers occurred in a groups similar to those of the bast. Xylem parenchyma were of two kinds, those associated with gelatinous fibres having thick pitted walls and the remaining with thin walls and lacking pits. The vessels were occurred mostly in a groups of 2-3 and have thick pitted walls and contained 3-5 rows of transversely oblique bordered pits. Vessels with annular thickening were also present. The cavities of some of the vessels present in the center are filled with reddish-brown contents. The medullary rays are almost uniform in size and 3 to 4 cell wide. Their cells are thick walled, pitted, radially elongate rectangular and fully loaded with starch grains of various sizes and prismatic crystals of calcium oxalate was quite characteristic and measured up to 36 µm in length. Starch grains which are mostly single, spherical, oval or elliptical or muller shaped , dimensions varying from 3 to 15 µm in length but sometimes attaining up to 21 µm. The starch grains with 2 components are few and 3 to 6 components are rare.

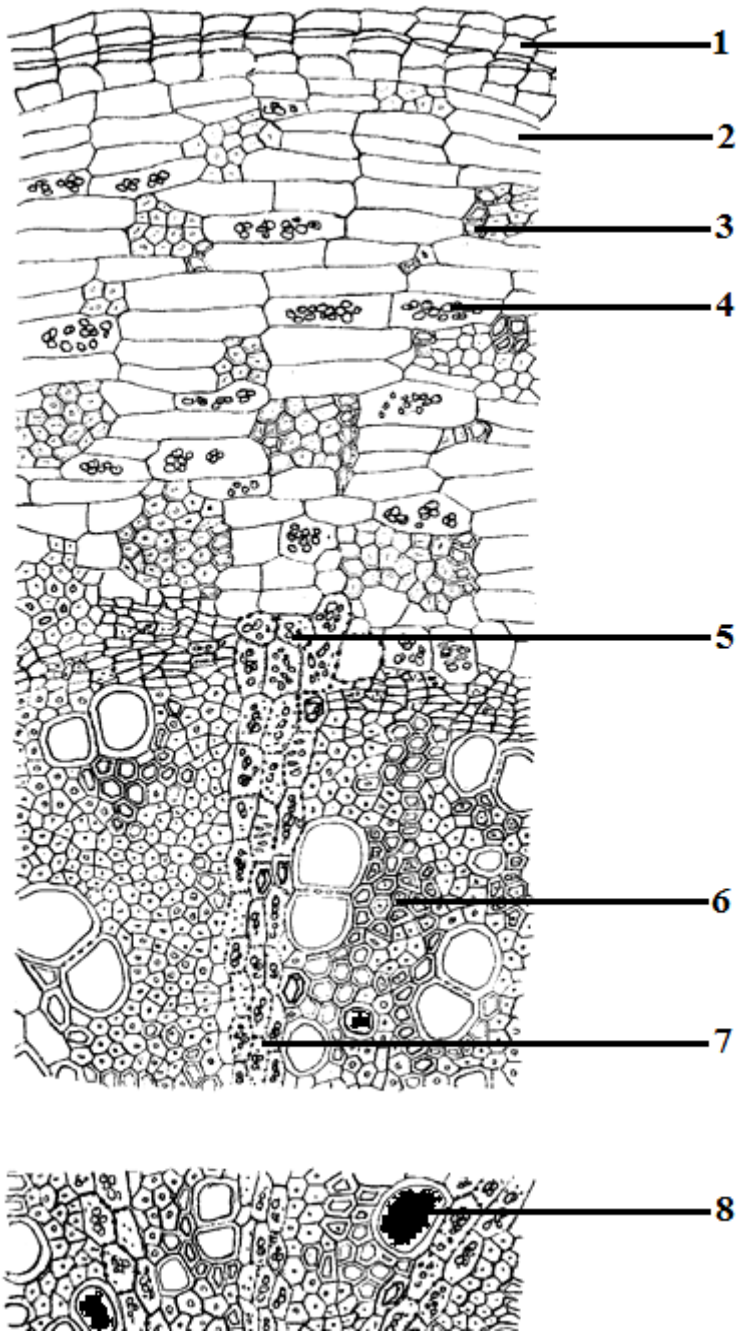


Fig.103.*Taverniera cuneifolia* root, T.S: 1.Cork, 2.Cortex, 3.Prismatic crystals, 4.Starch grains, 5.Phloem rays, 6.Gelatinous fibre 7.Xylem rays, 8.Vessels with reddish-brown contents.

Root : T.L.S (Fig.104)

The cork cells were many layered. Parenchyma cells in the cortical region were thin walled adhering to which were crystal fibres containing prismatic crystals. Thick walled bast fibers with narrow lumen appeared straight. Phloem rays, cells of which contained starch grains. The parenchymatous cells of the secondary phloem were thin walled. The xylem rays which were thick walled appeared multiseriate and contained starch grains. The vessels and tracheids contained bordered pits. Vessels were having straight end walls.

Root : R.L.S (Fig.105)

The gelatinous fibres were straight. The rays were upright and squar to rectangular in shape and each cell was filled with starch grains. Vessels showed the presence of boarded pits and few were annular thickened also .

Root : Powder study (Fig.106)

The powder was characterized by the presence of groups of light brown thick walled cork cells, cortical cells with starch , bast and wood fibers adhering to which were crystal fibers, containing prismatic crystals of calcium oxalate, thick walled fibers with narrow lumen and blunt tips, thick walled ray parenchyma cells with simple pits and having starch grains and boarded pitted vessels.

Distinguishing features

Phytochemical markers

1. Ferulic acid.
2. *o*-Coumaric acid.
3. Melilotic acid.
4. *p*-Hydroxy benzoic acid.

Pharmacognostic markers

1. Light brown thick walled cork cells.
2. Starch grains.
3. Prismatic crystals.
4. Crystal fibres.
5. Thick walled fibers with narrow lumen and blunt tips.
6. Thick walled pitted ray parenchyma.

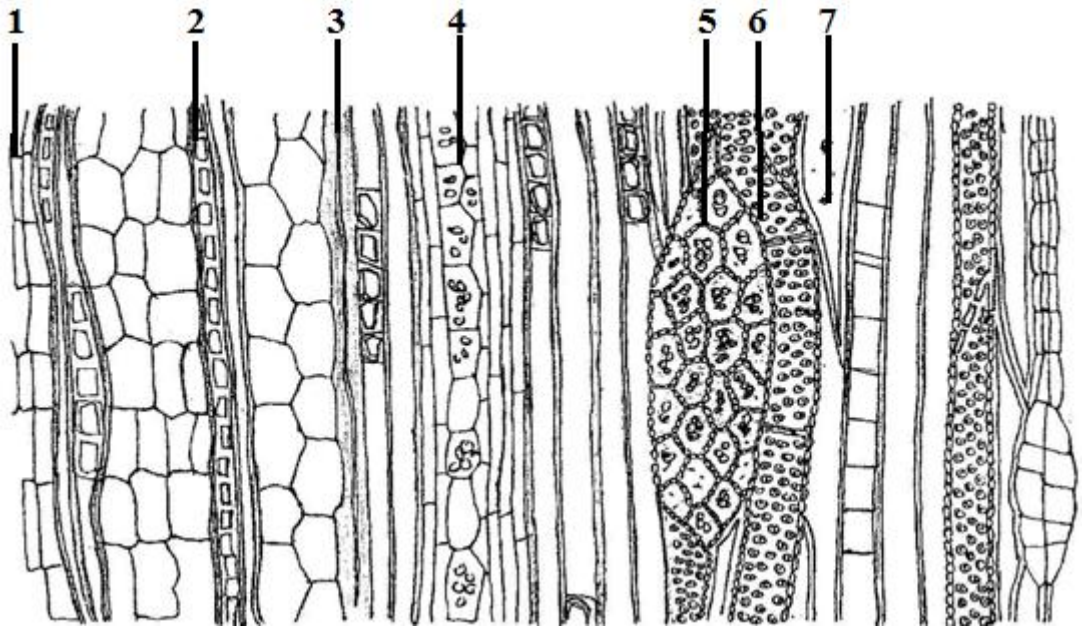


Fig.104.*Taverniera cuneifolia* root, T.L.S:1. Cork, 2. Crystal fibre, 3.Bast fibre, 4.Phloem rays,5. Xylem rays.6. Vessels with alternate bordered pits, 7. Fiber trachieds.

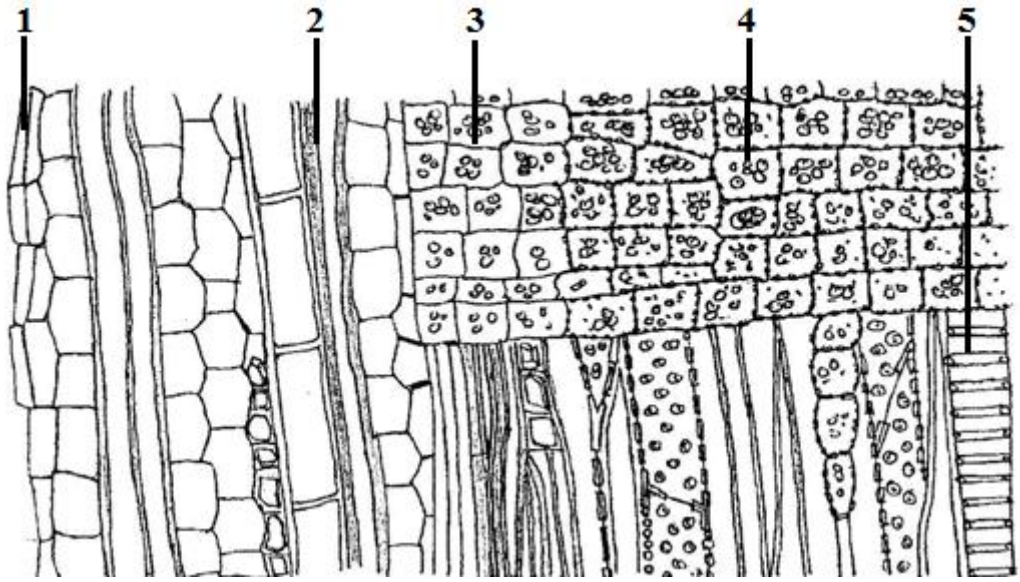


Fig.105. *Taverniera cuneifolia* root, R.L.S:1. Cork, 2. Gelatinous fibre, 3. Phloem rays with starch grains,4. Xylem rays, 5.Annular thickened vessels.

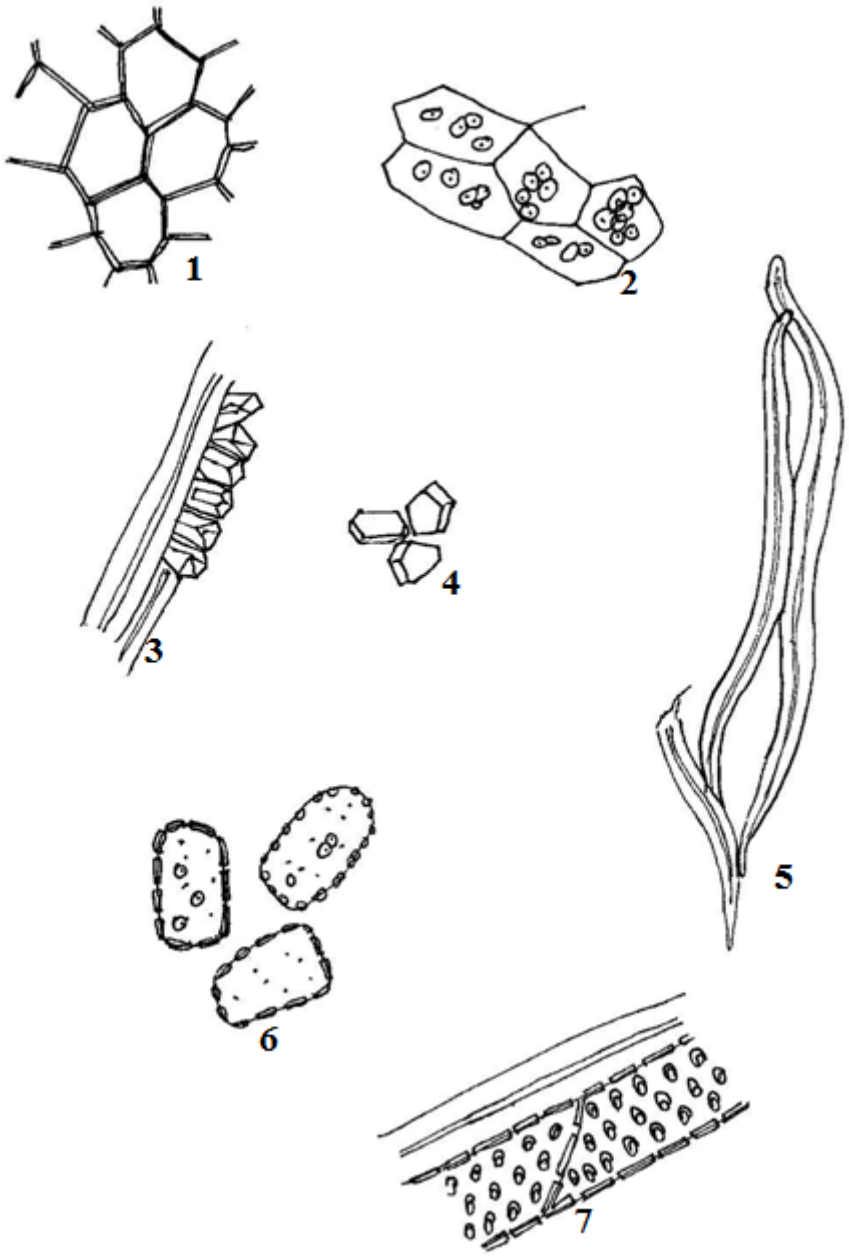


Fig.106.*Taverniera cuneifolia* root, Powder study: 1.Cork cells, 2.Parenchyma with starch grains, 3.Crystal fibre, 4.Prismatic crystals 5.Thick walled fibers with narrow lumen and blunt tips,6. Ray parenchyma, 7. Boarded pitted vessels.

Physico-chemical analysis:

Table 18 : Values obtained for the proximate analysis.

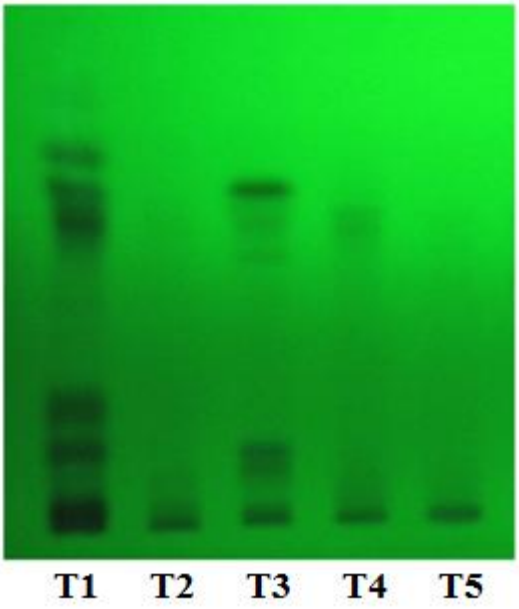
Sr.No.	Parameter	Mean ± SD (%)*			Average (%)
		Summer	Monsoon	Winter	
1.	Total Ash Content	5.71±0.36	5.86±0.41	5.79±0.39	5.79
2.	Acid Insoluble Ash content	1.08±0.08	0.99±0.06	1.06±0.04	1.04
3.	Alcohol soluble extractives	16.01±0.63	16.87±0.53	16.03±0.61	16.30
4.	Water soluble extractives	18.80±0.46	19.02±0.29	18.73±0.42	18.85

*Each value is a mean of 3 readings.

5.f. HPTLC fingerprinting and Physo-chemical analysis of *Glycyrrhiza glabra* and its substitutes/adulterants

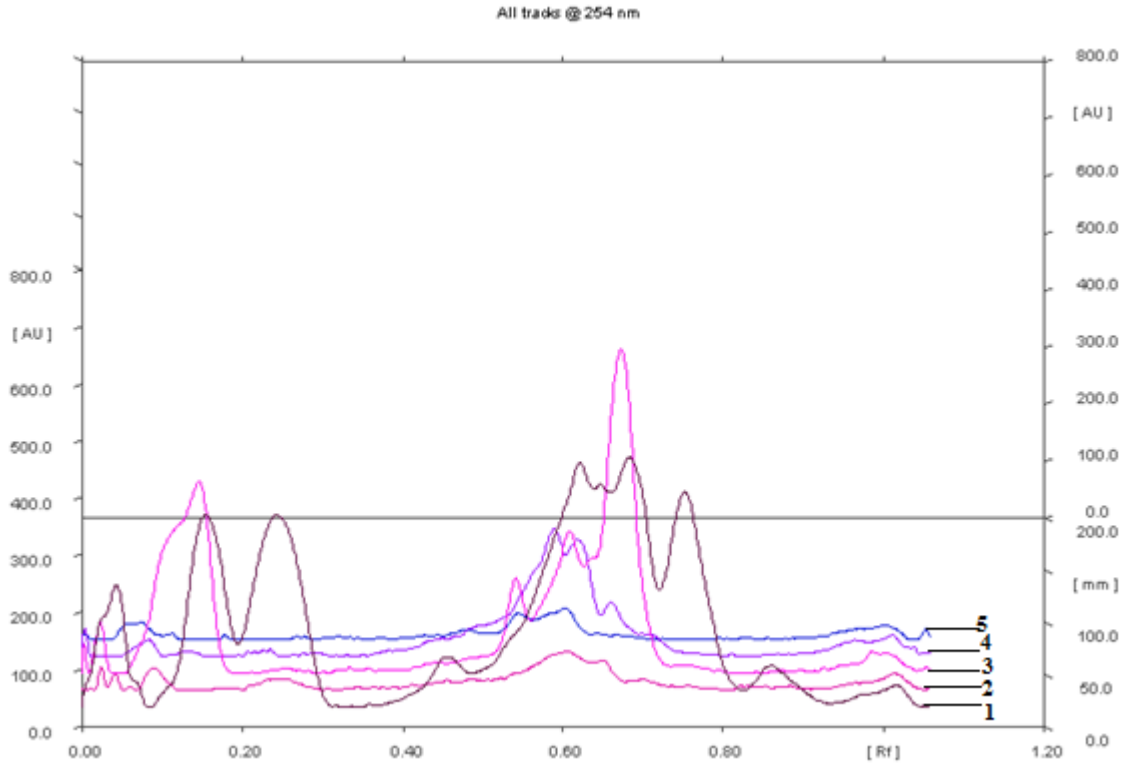
HPTLC fingerprinting

Figure 107.a : HPTLC chromatogram of *Glycyrrhiza glabra* and its substitutes/adulterants. (UV 254 nm).



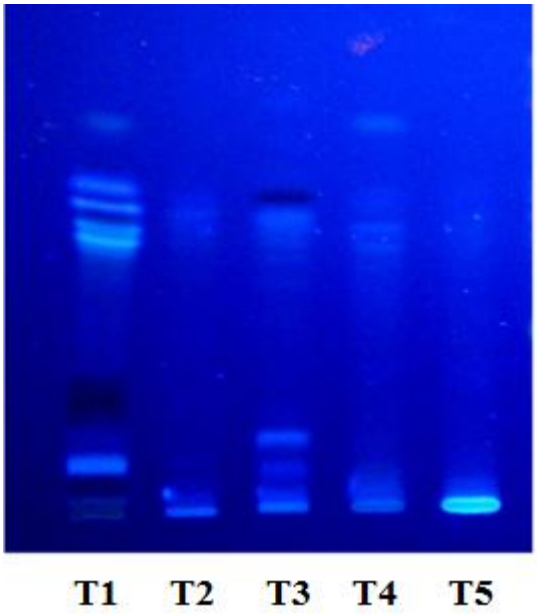
(a). T1-*Glycyrrhiza glabra*, T2-*Taverniera cuneifolia*, T3-*Abrus precatorius*, T4-*Alyscicarpus longifolius*, T5-*Maerua arenaria*.

Figure 107.b: HPTLC chromatogram of *Glycyrrhiza glabra* and its substitutes/adulterants. (UV 254 nm).



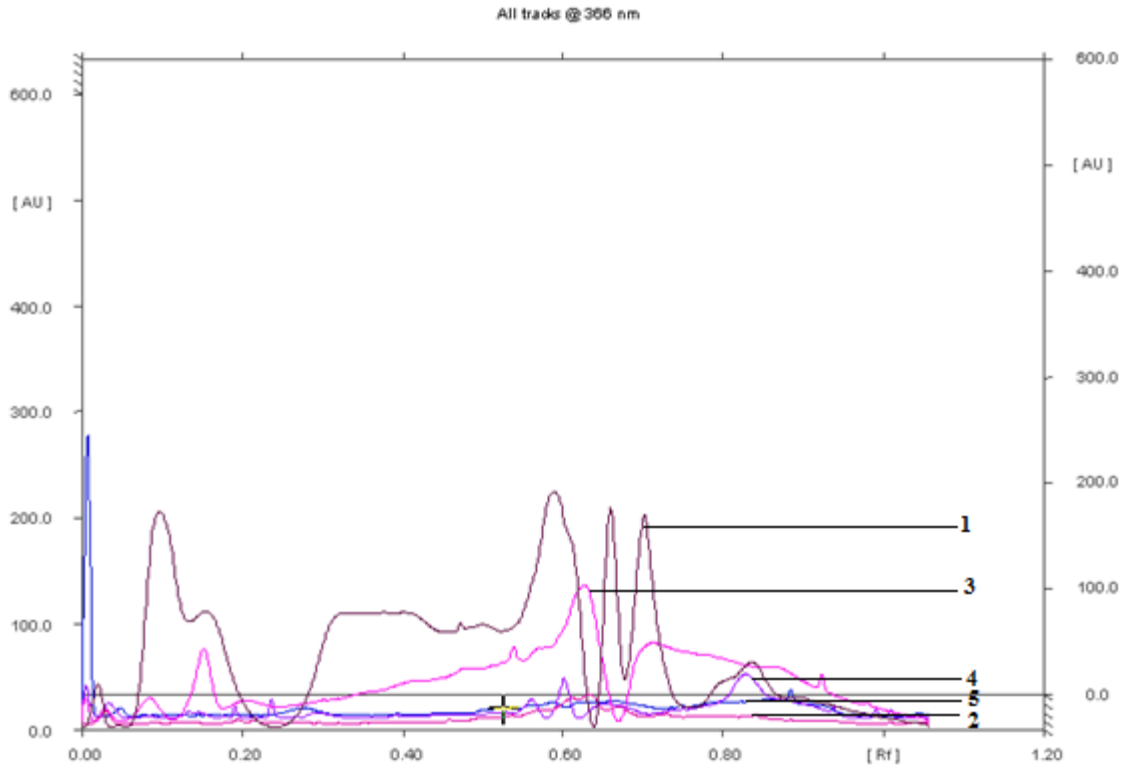
(b).1-*Glycyrrhiza glabra*, 2-*Taverniera cuneifolia*, 3-*Abrus precatorius*, 4-*Alyscicarpus longifolius*, 5-*Maerua arenaria* .

Figure 108.a : HPTLC chromatogram of *Glycyrrhiza glabra* and its substitutes/adulterants (UV 366 nm).



(a). T1-*Glycyrrhiza glabra*, T2-*Taverniera cuneifolia*, T3-*Abrus precatorius*, T4-*Alyscicarpus longifolius*, T5-*Maerua arenaria*.

Figure 108.b: HPTLC chromatogram of *Glycyrrhiza glabra* and its substitutes/adulterants (UV 366 nm).



(b). 1-*Glycyrrhiza glabra*, 2-*Taverniera cuneifolia*, 3-*Abrus precatorius*, 4-*Alyscicarpus longifolius*, 5-*Maerua arenaria* .

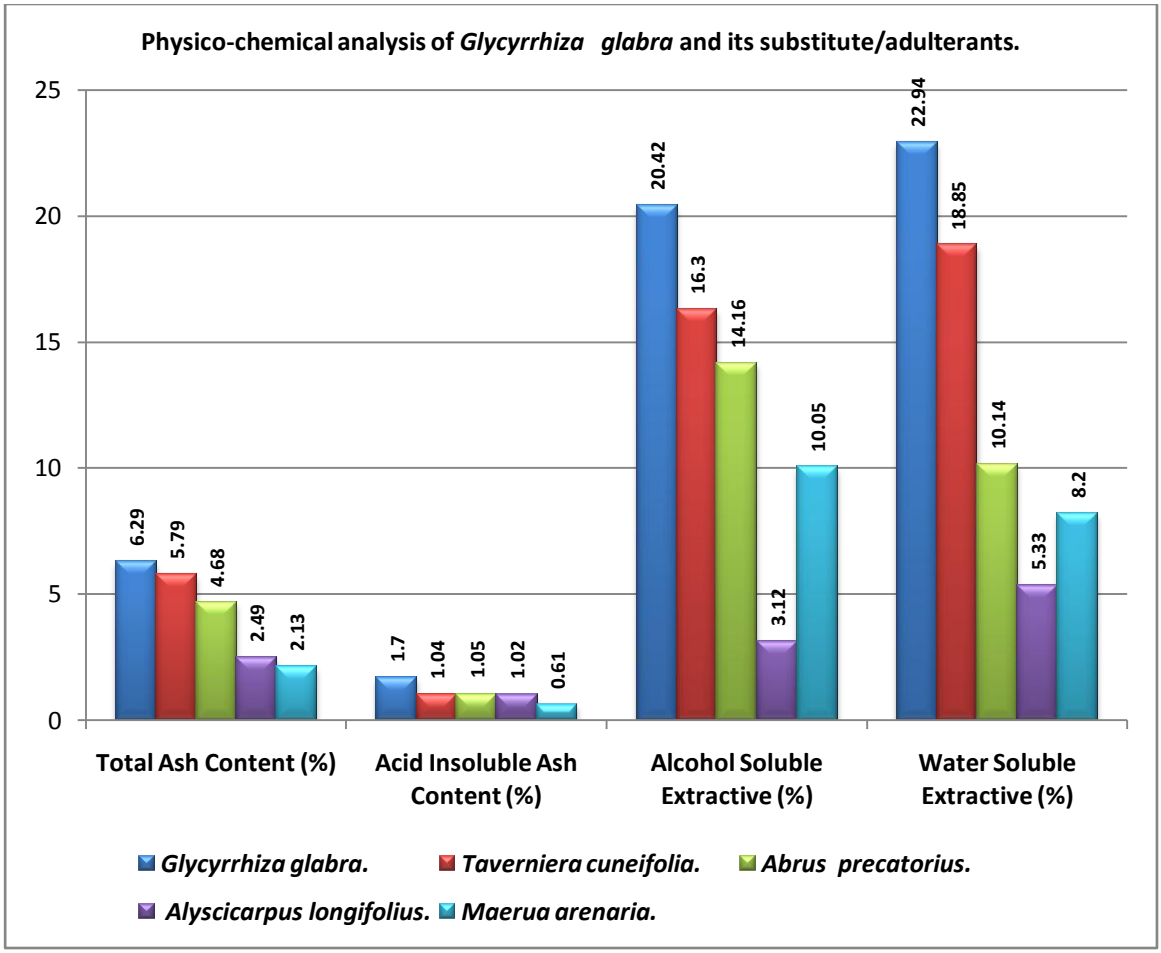
HPTLC profile of *Glycyrrhiza glabra* showed the presence of 10 peaks in both observed under UV 254 nm (figure 107.b)and 366 nm (figure 108.b). There were 5 major peaks at R_f 0.10, R_f 0.33 , R_f 0.40, R_f 0.59 and R_f 0.70 found under UV 254 and peaks at R_f 0.15, R_f 0.24 , R_f 0.62, R_f 0.68 and R_f 0.75 under 366 nm. The *Taverniera cuneifolia* showed the presence of 9 peaks, *Abrus precatorius* and *Alysicarpus longifolius* 8 peaks and *Maerua arenaria* 7 peaks when observed under UV 254 (figure-107.b)while under UV366 nm(figure 108.b); *Taverniera cuneifolia, Abrus precatorius, Alysicarpus longifolius* and *Maerua arenaria* showed the presence of 3,11,7 and 2 peaks respectively.

HPTLC profile of *G. glabra* and its substitutes/adulterants observed under UV 254 nm showed that *T. cuneifolia* was similar in 3 peaks but differed in 6 peaks. Both *Abrus precatorius* and *Alysicarpus longifolius* were similar in 1 peak but differed in 7 peaks, while *Maerua arenaria* was not show any peak similar to that of *G. glabra* but differed in having 7 peaks.

HPTLC profile of *G. glabra* and its substitutes/adulterants observed under UV 366 nm showed that both *Abrus precatorius* and *Alysicarpus longifolius* were similar in 1 peak but *Abrus precatorius* differed in 10 peaks and *A. longifolius* in 6 peaks while *T. cuneifolia* and *M.arenaria* did not showe any peak similar to that of *G.. glabra but T. cuneifolia* differed in 3 and *M. arenaria* in 2 peaks.

Physico-chemical analysis

Physico-chemical analysis of *Glycyrrhiza glabra* and its substitutes/adulterants



Total ash content

Total Ash Content of *Glycyrrhiza glabra* (6.29 %) along the material collected in different season does not show significant variation (Table-14) while the closest value to the substitute/adulterant in descending order is 5.79% (*Taverniera cuneifolia*), 4.68% (*Abrus precatorius*), 2.49%(*Alyscicarpus longifolius*) and 2.13% (*Maerua arenaria*).

Acid insoluble ash content

Acid insoluble ash content of *Glycyrrhiza glabra* (1.70 %) along the material collected in different season does not show significant variation (Table-14) while the closest value to the substitute/adulterant in descending order is 1.05% (*Abrus precatorius*),1.04% (*Taverniera cuneifolia*), 1.02% (*Alyscicarpus longifolius*) and 0.61% (*Maerua arenaria*).

Amongst the substitutes/adulterants of *Glycyrrhiza glabra*, the *Taverniera cuneifolia* showed the closest value of total ash content which showed that the *T.cuneifolia* was more close to *G. glabra*as compared to other substitutes/adulterants of *G. glabra*.

Alcohol soluble extractive

Alcohol soluble extractive value of *Glycyrrhiza glabra* (20.42%) along the material collected in different season does not show significant variation (Table-14) while the closest value to the substitute/adulterant was of *Taverniera cuneifolia* (16.3%).The values of *Abrus precatorius*, *Maerua arenaria* and *Alysicarpus longifolius* was found to be 14.16%,10.05% and 3.12% respectively.

Water soluble extractive

Water soluble extractive value of *Glycyrrhiza glabra* (22.94 %) along the material collected in different season does not show significant variation (Table-14) while the closest value to the substitute/adulterant in descending order is 18.85% (*Taverniera cuneifolia*),10.14% (*Abrus precatorius*), 8.14% (*Maerua arenaria*) and 5.33% (*Alyscicarpus longifolius*).

Amongst all substitutes/adulterants of *Glycyrrhiza glabra*, the *Taverniera cuneifolia* showed the extractive values close to the *G. glabra* while the extractive values of other substitutes/adulterants were almost half or less than half of *G. glabra* which reflect that the *T. cuneifolia* could better substitute as compared to other substitutes/adulterants.

Chapter 6

6.a.*Polygala senega* L. (Polygalaceae)

Synonyms: *Polygala rosea* Steud ; *Senega officinalis* Spach.

Vernacular names:

English : Senega Root, Snake Root.

Distribution and habitat

A small plant widely distributed over the United States and the southern parts of Canada (Wallis, 1960).

Morphological features

These are herbs or shrubs with upright, herbaceous to woody stems often branching profusely, the branches occasionally becoming geotropic or subterranean and bearing cleistogamous flowers. The leaves are simple, often lanceolate or linear, exstipulate, alternate. The inflorescence is a raceme or spike. Flowers are irregular, hermaphroditie with 5 distinct sepals, the 2 lateral ones being large and petaloid; 5 petals of which the two lateral are wanting or rudimentary and the anterior large and boat-shaped; 8 stamens; and a bicarpellate pistil. The fruit is a 2-celled capsule., rarely a drupe or samara. Pollen grains are barrel-shaped(Youngken,1951).

Medicinal uses:

Ethanolic extract of *Polygala senega* is used as an expectorant to treat cough, sore throat, bronchitis, and asthma (Lacaille *et al.*,2005, Kindscher,1992) and as an antihypoglycemic agent (Kako *et al.*,1996). The saponins of *P. senega* are used as vaccine adjuvants to increase specific immune responses (Katselis *et al.*,2007) and it is remedy for snake-bite(Wallis,1960).

Previous Phytochemical reports

Senegin,polygalic acids,methyl salicylate and fixed oil are the compounds reported(Wallis, 1960).

Previous pharmacognostic reports

Only the T.S of the root has been studied (Anon. 2004). So a detailed study with T.L.S and R.L.S is conducted on root of the plant.

Materials and methods

The plant material has been procured from authentic vender, Mumbai. Phytochemical analysis of roots of the plant for their secondary metabolites such as alkaloids, flavonoids, phenolics, steroids, quinones, mucilages and other compounds were done by standard methods of chromatography and spectrophotometry described in chapter 2. Pharmacognostic, HPTLC finger printing and physico-chemical studies were done by using standard methods described in chapter 2.

Results

Phytochemistry

The roots of the plant were found to contain saponin, steroids, vanillic and syringic acids while flavonoids were found to be absent. Mucilage amounted to 5.02 % consisting of arabinose, ribose, rahmnose and xylose.

Pharmacognosy

Macroscopic characters (Fig.109)

The roots were thick, tortuous, vertical mostly with a knotty crown at the top which gradually tapers at the end. Surface longitudinally wrinkled with few transverse scares and yellowish brown to light brown in colour; odour particular. fracture short and somewhat splintery in the centre.

Fig.109. *Polygala senega*, root

Microscopic characters

Root : T.S (Fig. 110)

The root possessed a few layered cork where the cells were thin and rectangular and were yellowish brown followed by a single layered phellogen. Phelloderm consisted of two to five layers of oblong parenchymatous cells of which some of them became collenchymatous and filled with oil globules while others were with light yellow contents. . Narrow zone of secondary cortex was made up of three to four rows of compactly arranged parenchymatous cells traversed by medullary rays. The medullary rays were V- shaped very broad towards the outer side and cells contained oil globules. The active phloem was 5-6 layers of thin walled cells. The xylem composed of mostly tracheids and vessels along with many thick walled wood parenchyma. The primary xylem was diarch. Vessels were many, scattered.

Root : T.L.S (Fig.111)

The cells of the cork were rectangular and contained yellowish- brown inclusions. Below cork were radial rows of rectangular thick walled collenchyma cells contained light yellow oil globules. The phloem rays were thin walled and polygonal containing light yellow coloured content and oil globules. The vessels were narrow, with 3-4 alternate rows of bordered pits. The xylem rays were spindle shaped and contained light yellow coloured content and oil globules.

Root : R.L.S (Fig.112)

The phloem ray cells were erect, polygonal and were filled with light yellow coloured content and oil globules. Between the cells of phloem and xylem rays there were the single upright column of the cambium was found. Xylem rays were heterogeneous square to hexagonal and contained oil deposits. Primary xylem had tracheids with spiral thickening.

Root : Powder study (Fig.113)

The components present in the powder were yellowish-brown cork cells, collenchyma with oil droplets, ray parenchyma with oil droplets, lignified parenchyma, bordered pitted vessel and tracheids.

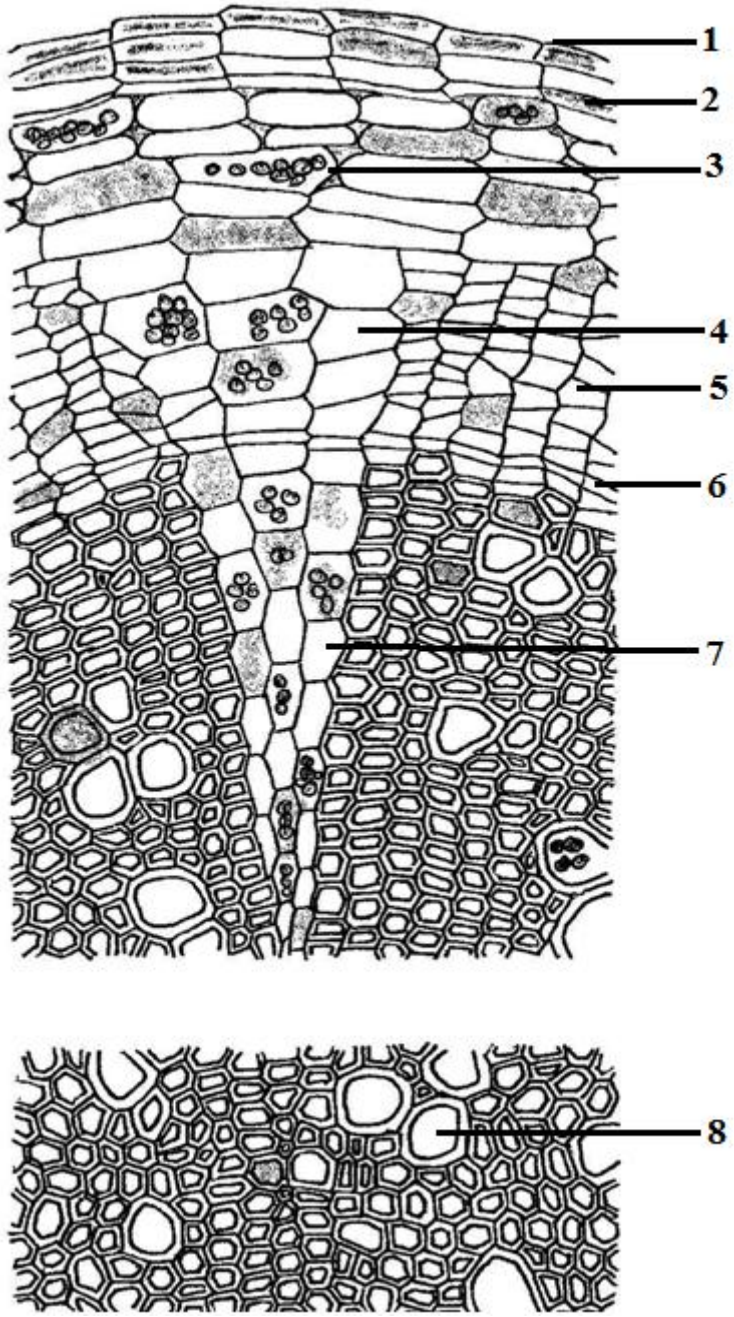


Fig.110. ***Polygala senega*** **root, T.S**: 1. Cork, 2. Phellogen, 3. Cortex,4. Oil droplets, 5. Broad V-shaped medullary ray, 6. Phloem 7. Cambium, 8.Xylem rays, 9. Vessels.

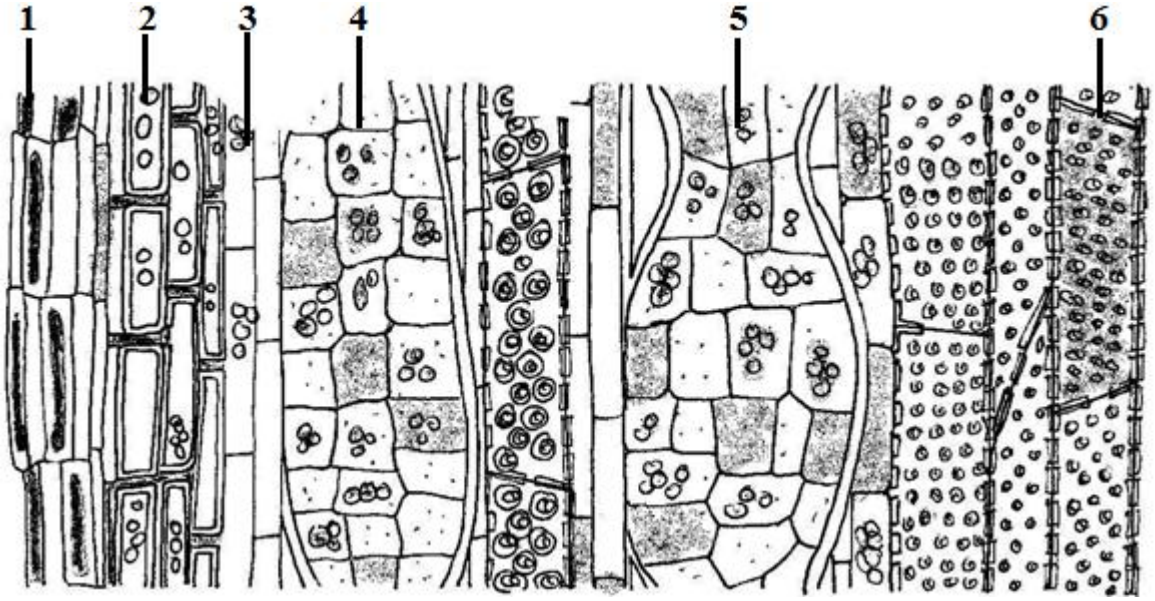


Fig.111. ***Polygala senega*** **root, T.L.S**:1. Cork, 2 Collenchyma with oil droplets, 3.Phloem parenchyma, 4. Phloem rays, 5. Xylem rays, 6. Vessels with alternate bordered pits.

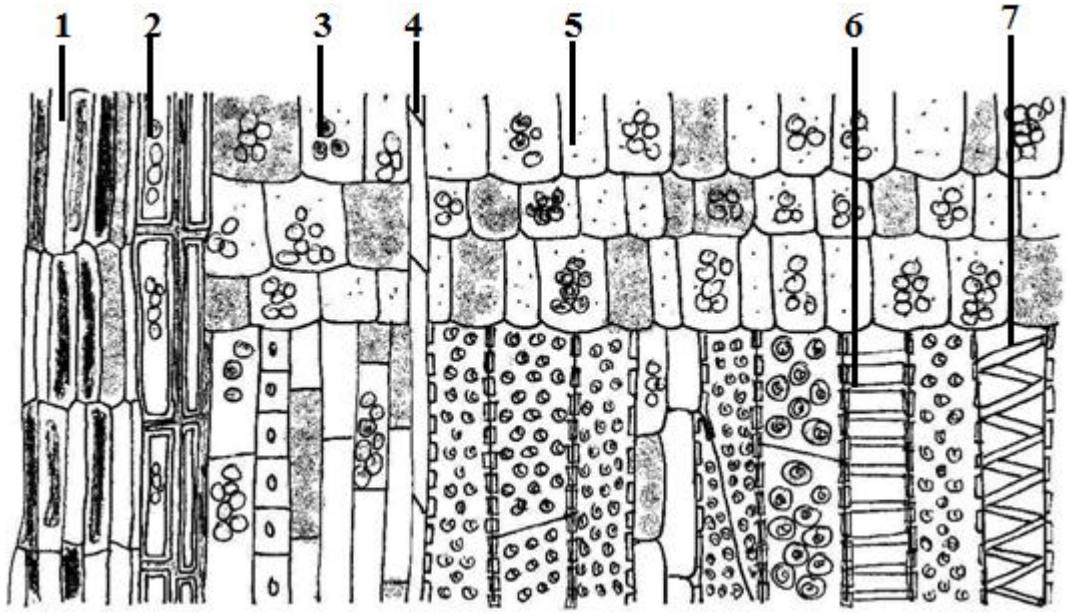


Fig.112. ***Polygala senega*** **root, R.L.S**:1. Cork, 2. Collenchyma with oil droplets, 3.Phloem rays, 4. Cambium, 5. Xylem rays, 6.Annular thickened vessel, 7.Vessels with spiral thickening.

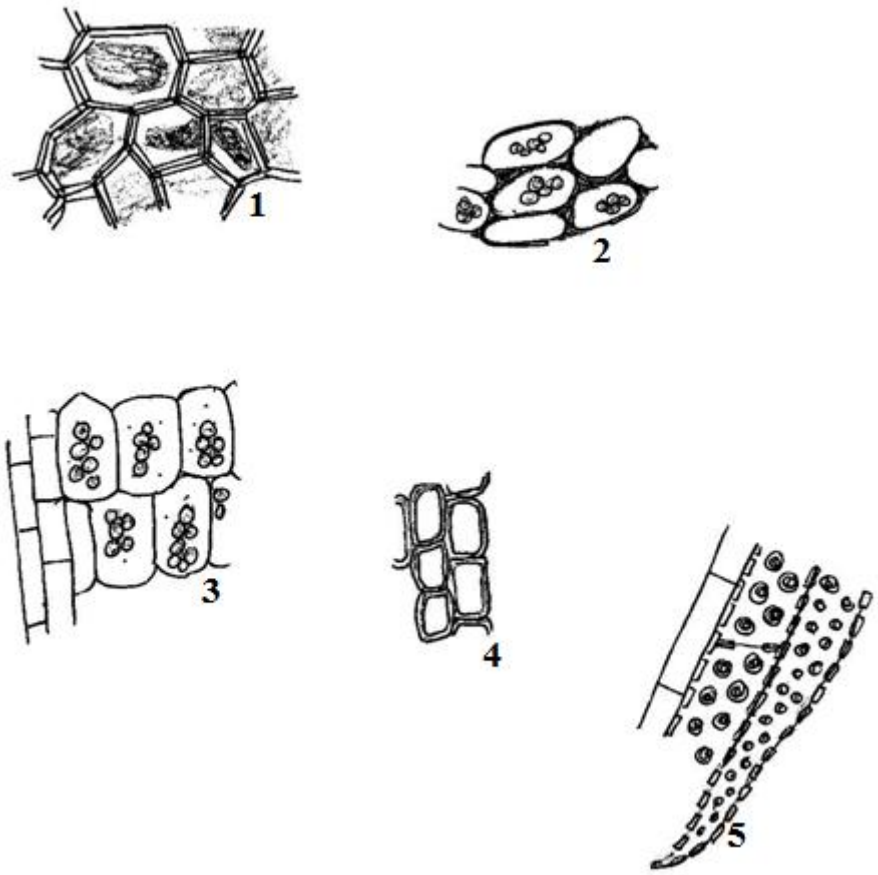


Fig.113. ***Polygala senega*** **root,Powder study:** 1.Yellowish-brown cork cells, 2.Collenchyma with oil droplets, 3. Ray parenchyma with oil droplets, 4. Thick walled wood parenchyma,5.Bordered pitted vessel and tracheids.

Distinguishing features

Phytochemical markers

1. Vanillic acid.
2. Syringic acid.
3. Rhamnose.
4. Xylose.
5. Arabinose .
6. Ribose.
7. Absence of flavonoids.

Pharmacognostic markers

1. Yellowish-brown cork cells.
2. Collenchyma with oil droplets.
3. Medullary rays were broad forming V- shape.
4. Ray parenchyma with oil droplets.
5. Thick walled wood parenchyma.
6. Bordered pitted vessel and tracheids.

Physico-chemical analysis:

Table 19 : Values obtained for the proximate analysis.

Sr.No.	Parameter	Mean ± SD (%)*			Average (%)
		Summer	Monsoon	Winter	
1.	Total Ash Content	5.39±0.31	5.36±0.34	5.33±0.33	5.36
2.	Acid Insoluble Ash content	1.11±0.07	1.09±0.08	1.10±0.03	1.1
3.	Alcohol soluble extractives	18.62±0.38	18.89±0.66	18.17±0.49	18.56
4.	Water soluble extractives	28.80±0.41	29.02±0.29	28.77±0.38	28.86

*Each value is a mean of 3 readings.

6.b. *Acalypha indica* Linn. (Euphorbiaceae)

Synonyms : ***Acalypha canescens*** Wall.

Sanskrit names: Arittamanjarie, Rudra, Muktavarchas.

Vernacular names:

Bengal : Muktajhuri,Sveta-basanta, harita manjari.

English : Indian acalypha.

Gujarati : Vanchi Kanto.

Hind : Kuppu,Khokali, khokla, khokli, kuppi, kuppikhokli, kholi

Kannada : Kuppigida.

Konkani : Kunkmiphal.

Malayalam : Kuppamani.

Tamil : Kuppivaeni; Kuppaimeni.

Telugu : Kuppichettu, Harita-manjiri, Kuppinta,Muripindi.

Uriya : Indramaris.

Distribution and habitat

The plant is a common weed found in waste places, gardens and along the road sides throughout the hotter parts of India.

Morphological features:

An annual erect herb, 30-100 cm high, branches numerous, long, ascending, angular, finely pubescent. Leaves 2.5 - 7.5 cm long and 2-2.5 cm broad, ovate or rhomboid- ovate, acute or sub-obtuse, crenate- serrate, glabrous, thin, base cuneate, somewhat 3- nerved; petioles usually longer than the blade, slender; stipules minute. Flowers unisexual, in numerous lax, erect, elongate axillary spikes; the male flowers minute, terminal or axillary; the female flowers scattered, 3-7, surrounded by a short pendunculate, large, leafy, truncate, dentate, cuneiform, many-nerved bract, 6-8 mm in diameter. Ovary hispid. Capsules small, hispid, quite concealed by the bract, often only 1-seeded. Seeds ovoid, smooth, pale-brown.

Medicinal uses:

The plant is emetic, purgative, beneficial in cough, dyspnoea, fever, deranged kapha and vata. Fresh leaf extract with common salt is applied in eczema. It is used in gastrointestinal and respiratory affections (Raj and Singh 2000) and is a useful remedy for bronchitis, asthma and pneumonia. The plant extracts have significant antioxidant properties(Durga *et.al,* 2009). The methanolic extract showed analgesic and anti-inflammatory effect (Rahman *et.al,* 2010) .The water extract showed the

maximum zone of inhibition for *Staphylococcus aureus* and *Pseudomonas aeruginosa* (Raja *et al.*,2009). According to Siddha Materia Medica the leaf powder when given in the dose of 950 mg to 1300 mgs, cures respiratory diseases (Muthaliar 1988). The leaf juice when mixed with neem oil and applied to the inner part of children's tongue with the help of quill, induces vomiting and acts as expectorant. The root is employed as a cathartic. In small doses it is expectorant and nauseant and in large doses emetic(Datta and Mukharji 1950). Roots acts as a laxative and anthelmintic (With garlic) (Chopra *et al.*,1956;Watt,1972; Desai, 1975; Dymock *et al.*,1976;Bhandari,1977). The ethanol and aqueous extracts of root exhibited significant antioxidant activity (Balakrishnan *et al.*,2009; Durga *et al.*,2010).

Previous Phytochemical reports

The plant contained a cyanogenetic glucoside, acalyphine, two alkaloids, viz, acalyphine and triacetonamine, an essential oil n-octacosanol, quebrachitol, b-sitosterol acetate and tannin. Acalyphamide (as acetate), aurantia-mide and its acetate, succinimide calypho lacetate, 2-methyl anthraquinone, tri-O-methylellagic acid, b-sitosterol and its β -D-glucoside in leaves ; stigmasterol in root (Raj and Singh 2000) and four known kaempferol glycosides, mauritianin, clitorin, nicotiflorin and biorobin, have been isolated from the flowers and leaves (Nahrstedt *et al.*, 2006) and B-group vitamins were detected in the plant (Rao *et al.*,1982).

Previous pharmacognostic reports

Only the T.S of the root has been studied (Raj and Singh 2000; Datta and Mukharji 1950).

Materials and methods

The plant material has been collected from Vadodara, Gujarat. Phytochemical analysis of roots of the plant for their secondary metabolites such as alkaloids, flavonoids, phenolics, steroids, quinones, mucilages and other compounds were done by standard methods of chromatography and spectrophotometry described in Chapter 2. Pharmacognostic, HPTLC finger printing and physico-chemical studies were done by using standard methods described in Chapter 2.

Results

Phytochemistry

The acalyphine alkaloid which has been reported earlier, was located in the present investigation in roots in traces. The flavonoids were found to be absent. The

phenolic acids present were vanillic, syringic, and *cis* and *trans* ferulic acids. The mucilage amounted to 2.8% consisting of xylose and glucose.

Pharmacognosy

Macroscopic characters (Fig.114.)

The roots were vertical, woody, somewhat tortuous and of a pale buff colour; odor not very particular, taste slightly bitter. Fracture short.

Fig. 114. *Acalypha indica* Linn. root

Microscopic characters

Root : T. S (Fig. 115)

The T.S of the root was circular in outline. The cork consisted of 4 to 7 rows of thin walled, tangentially elongated cells of which the outermost one or two rows of cells were slightly ruptured. The secondary cortex was narrow zone consisting of 5 to 7 rows of isodiametric, thin walled parenchyma and arranged compactly. The cells of outer rows of cortex were narrow and tangentially elongated. Many of the cortical cells were filled with rosette crystals and simple spherical starch grains. Occasionally thick walled broad lumened sclerenchymatous fibres with distinct striations were also found in this region. The phloem consisted of usual phloem elements and many of them filled with rosette crystals and simple spherical starch grains. Phloem rays were uni- to bi-seriate and the cells were thin walled and contained starch grains. Wood consisted of vessels, tracheids, fibres and few wood parenchyma. Fibres were thick

walled and of various sizes and shapes with wavy margins. Xylem rays were thick walled containing starch grains. Starch grains were mostly simple and spherical or ovoid in shape. Vessels were simple and bordered pitted, mostly occurred singly but few were in groups of two and found associated with fibre tracheids. Spiral and scalariform thickened vessels were also common.

Root : T.L.S (Fig.116)

Cork cells appeared rectangular with thin walls.The sclerenchymatous fibres in the middle of the cortex were curved and spindle shaped. Some phloem parenchyma showed the presence of starch and rosette crystals. The cells of the rays were thin walled and contained starch grains. Xylem rays were comparatively thick walled, spindle shaped biseriate, simple pitted and contained starch grains. Tracheids contained multiseriate bordered pits. The bordered pits in vessels were arranged loosely in 4-6 rows.

Root : R.L.S (Fig.117)

The phloem ray cells appeared rectangular. They were thin walled and filled with starch grains in each cell. The pits on the wall were of simple type. The vessels showed 3-4 rows of bordered pits. The Primary xylem had spiral and annular type of thickenings.

Powder study (Fig. 118)

The components present in the powder were fragments of cork cells, rosette crystals of calcium oxalate, starch grains, thick walled broad lumened sclerenchymatous fibres with distinct striations, scalariform vessels, fibres with distinct lumen and wavy margins.

Distinguishing features

(I) Pharmacognostic markers

1. Rosette crystals.
2. Starch grains.
3. Thick walled broad lumened sclerenchymatous fibres with distinct striations.
4. Scalariform vessels.

(II) Phytochemical markers

1. *p*- Coumaric acid.
2. Ferulic (*cis*- and *trans*-isomers) acid.
3. Absence of flavonoids.

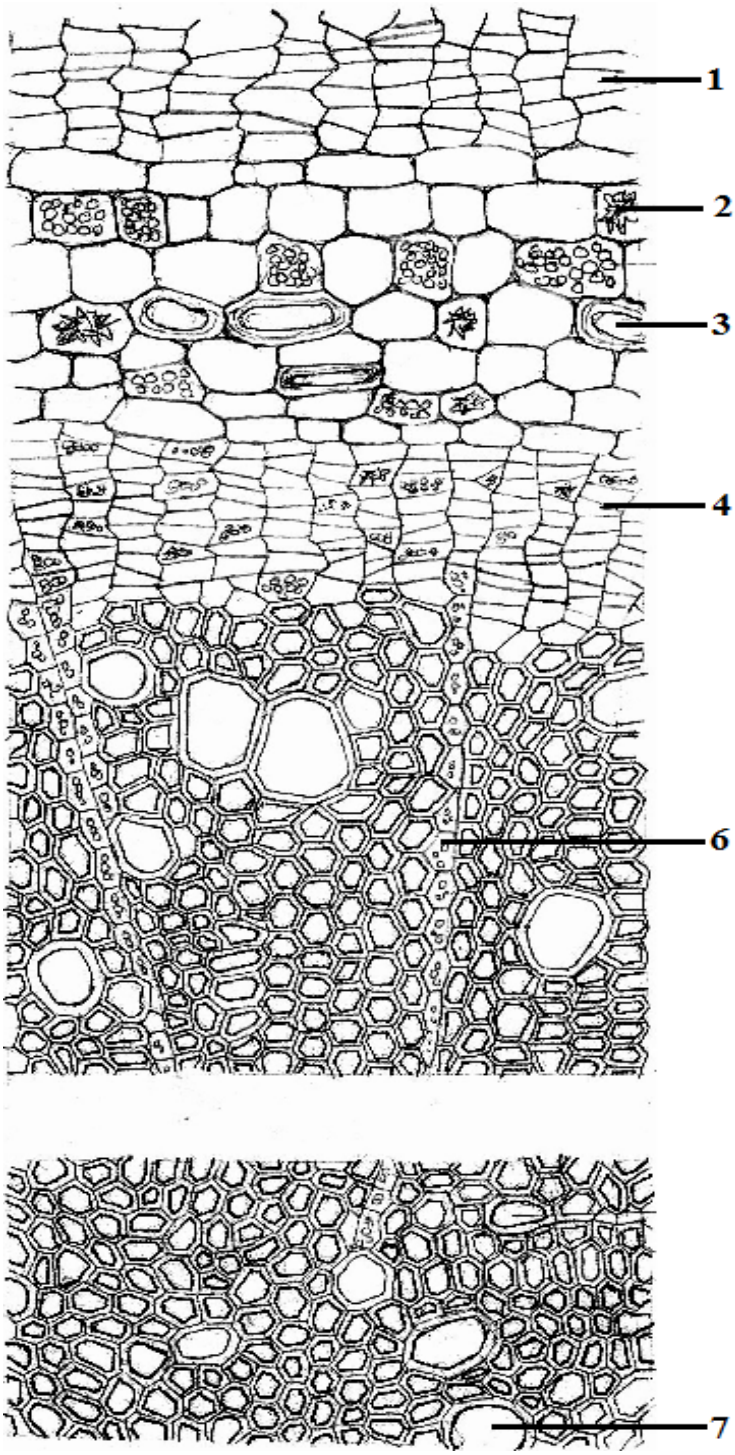


Fig.115. ***Acalypha indica*** **Linn. root, T.S**: 1.Cork, 2. Rosette crystal, 3.Parenchyma containing starch grains, 4. Sclerenchymatous fibre, 5. Phloem, 6. Xylem rays with starch grains, 7. Fibre tracheids, 8.Vessels.

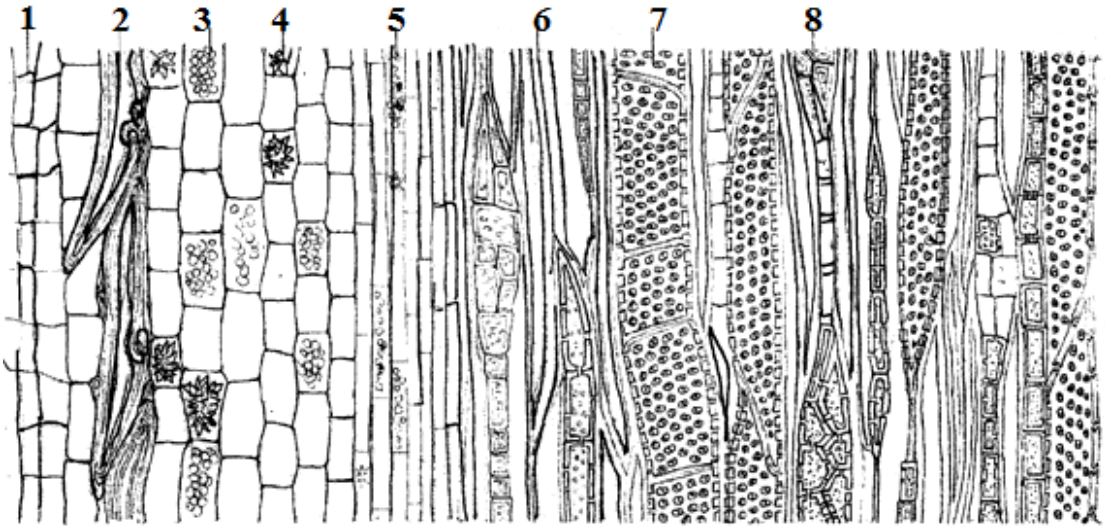


Fig.116. *Acalypha indica* root, T.L.S:1.Cork, 2.Sclerenchymatous fibre, 3.Starch grains, 4.Rosette crystal, 5. Phloem rays with starch grains, 6.Fibre tracheids, 7.Vessels, 8. Xylem rays.

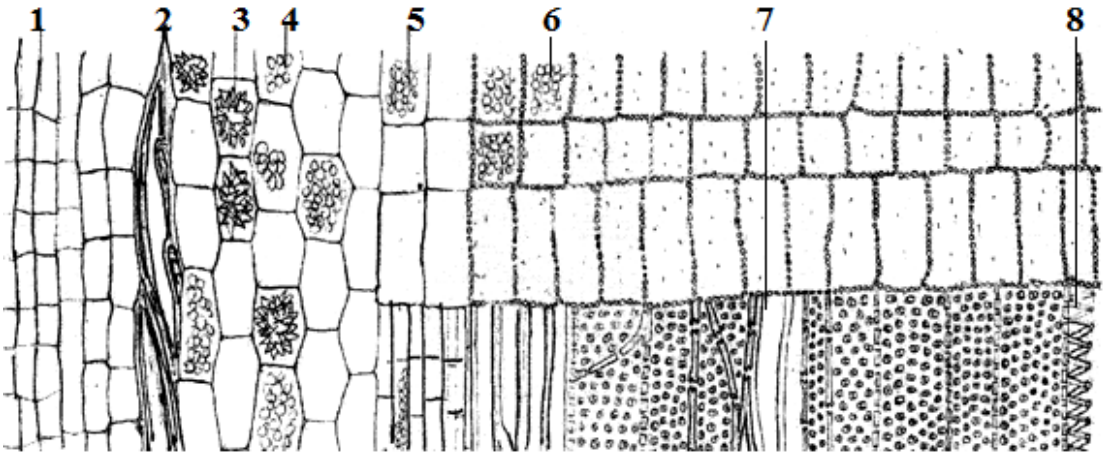


Fig.117.*Acalypha indica* root, R.L.S: 1.Cork, 2. Sclerenchymatous fiber, 3. Rosette crystal, 4. Starch grains, 5.Phloem rays with starch grains, 6. Xylem rays with starch grains, 7. Fibre tracheids, 8. Spiral vessel.

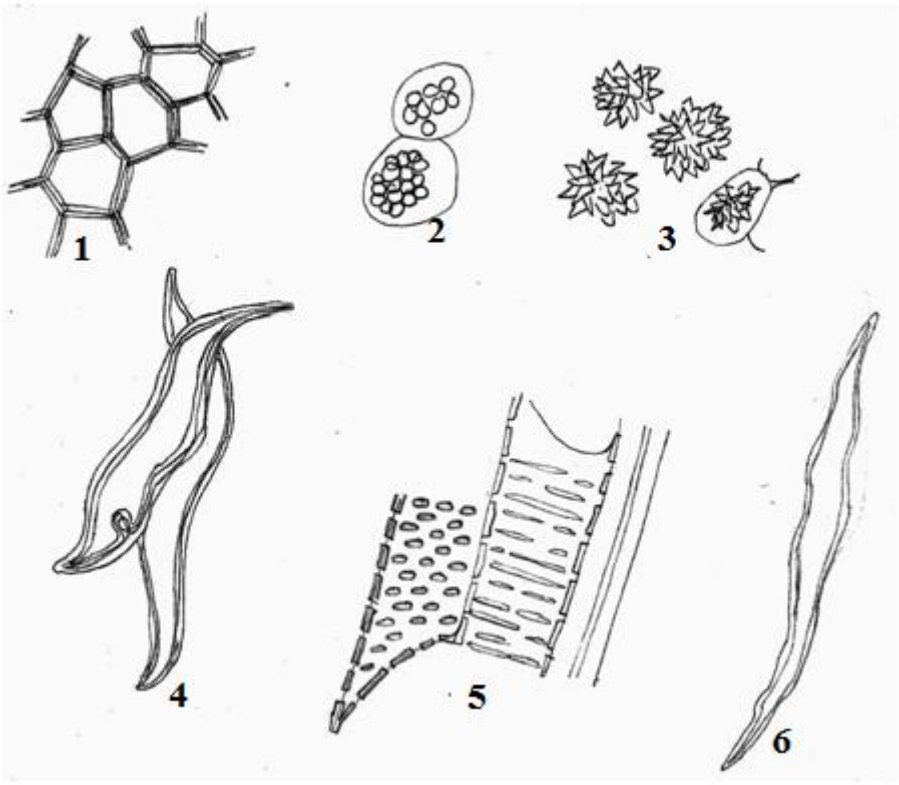


Fig.118. ***Acalypha indica*** **root, Powder study**: 1.Cork, 2. Parenchyma containing starch grains, 3. Rosette crystals, 4. Thick walled Sclerenchymatous fibres with distinct striations, 5. Scalariform vessels, 6. Fibre with wavy walls.

Physico-chemical analysis:

Table 20: Values obtained for the proximate analysis.

Sr.No.	Parameter	Mean ± SD (%)*			Average (%)
		Summer	Monsoon	Winter	
1.	Total Ash Content	02.68±0.29	02.93±0.31	02.35±0.36	2.65
2.	Acid Insoluble Ash content	0.97±0.35	0.97±0.84	0.97±0.	0.97
3.	Alcohol soluble extractive	01.51±0.11	01.31±0.17	01.52±0.15	1.45
4.	Water soluble extractive	03.43±0.31	02.91±0.24	02.98±0.39	3.11

*Each value is a mean of 3 readings

6.c.*Adhatoda vasica* L. Nees (Acanthaceae)

Synonyms: ***Adhatoda zeylanica*** Medic. A. (L.), ***Justicia adhatoda*** L.

Sanskrit : Atarusa, Simhasya, Vasaka.

Vernacular names:

Assamese : Bahak, Titabahak, Vachaka.

Bengali : Bakas, Basak.

English : Malabar nut.

Gujrati : Aradusi, Ardusi, Araduso.

Hindi : Adoosa, Aduss, Arusa.

Kannada : Adusoye.

Kashmiri : Vasa.

Malayalam : Adalodakam, Adarooshaka.

Manipuri :Nongmangkha Angouba.

Marathi : Adulsa, Vasa.

Oriya : Basanga, Vasanga.

Punjabi : Arusa, Bhekar, Vansa, Vishuti.

Tamil : Adadodi, Kattumurungai, Vachai, Atatotai, Akacattamarai.

Telugu : Addasaramu.

Urdu : Adoosa, Adusa , Arusa , Burg Bansa.

Distribution and habitat

The plant is an evergreen shrub distributed throughout India upto an altitude of 1300 m. and also cultivated.

Morphological features

A dense shrub 3 m high with many long opposite ascending branches; stem with yellowish bark, terete, glabrous. Leaves upto 20 cm long, elliptic-lanceolate, acuminate, and minutely puberulous when young, glabrous when mature, entire, dark-green above, paler beneath, base tapering; main nerves 10-12 pairs with reticulate venation between. Flowers in short dense axillary pedunculate spikes 8 cm long, towards the ends of the branches; peduncles shorter than the leaves; bracts elliptic, subacute, glabrous or nearly so, 5-7 nerved, closely reticulately veined; bracteoles oblong-lanceolate, acute, with ciliolate margins, 1-nerved, reticulately veined. Calyx

rather less than 1 cm long, glabrous or slightly pubescent, divided to the base; segments imbricate, oblong-lanceolate, acute, 3-nerved, reticulately veined. Corolla white, with a few irregular rose-colored bars in the throat, 3 cm long, pubescent outside; tube 1 cm long, the lower half cylindric, the upper half much laterally inflated; upper lip ovate-oblong, curved, obtuse, notched; lower lip as long as the upper, the lobes 1 cm deep, oblong, rounded, the middle lobe the broadest. Filaments hairy at the very base, long, stout, curved; lower anther-cells minutely apiculate at the base. Ovary pubescent; solid stalk flattened, 1 cm long. Seeds orbicular-oblong, tubercular-verrucose, glabrous.

Medicinal uses:

It is used as an herbal remedy for treating cold, cough, whooping cough and chronic bronchitis and asthma, as sedative expectorant, antispasmodic, anthelmintic and other pulmonary infections(Singh *et al.*,2011).It is also known for its antiarthritis, antiseptic, antimicrobial and antituberculosis properties (Dey, 1980). It is an important drug prescribed for malarial fever, fever caused by *pitta* and *kapha*, chronic fever, intrinsic hemorrhage, leprosy, skin diseases and piles (Soni, 2008).

Previous Phytochemical reports

The plant contains pyrroquinazoline alkaloids viz. vasicine, vasicol, vasicinone, peganine along with other minor constituents.Minor alkaloids include adhatonine vasicinol and vasicinolone. Flowers mainly yield of kaempferol and quercetin. A new moiety' 2 -4-dihydroxy chalcone-4-glucoside has been identified in the flowers. Four quinazoline alkaloids: vasicoline, adhatodine, vasicolinone and anisotine have been obtained from the leaves and vasicinone, vasicol have been isolated from the inflorescence. Sitosterol, ß -glucoside-galactose and deoxy vasicine have been isolated from the roots of the plant. Phytochemical investigations of leaves also yielded a quinazoline alkaloid identified as 1,2,3,9 tetrahydro-5-methoxypyrroloquinazoline-3-ol (Sayeed, 2009).

Previous pharmacognostic reports

Only the T.S of the root has been done (Anon.2004; Gupta *et al.*,2008) but study of T.L.S and R.L.S is remaining to be done. So a detailed study is conducted on root of the plant.

Materials and methods

The plant material has been collected from Vadodara, Gujarat. Phytochemical analysis of roots of the plant for their secondary metabolites such as alkaloids, flavonoids, phenolics, steroids, quinones, mucilages and other compounds were done by standard methods of chromatography and spectrophotometry described in Chapter 2. Pharmacognostic, HPTLC finger printing and physico-chemical studies were done by using standard methods described in Chapter 2.

Results

Phytochemistry

There was no flavonoid in the root. The phenolic acids located were vanillic acid, syringic acid along with trace amounts of *p*-coumaric acid, *p*-hydroxy benzoic acid and ferulic acid (*cis*- and *trans*-isomers). Mucilage amounted to 2.46 % consisting of xylose and galactose. The root also showed the presence of alkaloids and steroids while coumarins and saponins were found in good concentrations.

Pharmacognosy

Macroscopic characters (Fig.119)

The tap root was cylindrical and woody. The outer surface was yellowish brown in colour and showed longitudinal wrinkles. Fracture hard , Odour not distinct.

Fig.119. *Adhatoda vasica* root.

Microscopic characters

Root : T.S (Fig. 120)

The T.S of the root was circular in outline with a large central woody region and a thin outer bark. The cork consisted of 4 to 8 rows of thin walled, rectangular to tangentially elongated cells. Inner to the cork was the phellogen consisting of a single row of narrow thin walled tangentially elongated cells. The secondary cortex was consisting of five to ten rows of comparatively large polygonal thin walled parenchyma cells where the cells towards periphery were comparatively bigger than that of inner ones. Isolated groups of 2 to 3 stone cells were embedded towards the centre in the cortex. The stone cells were mostly squarish to rectangular and of narrow lumened. The cells of cortex were also showed the presence of scanty groups of small simple starch grains and were spherical or ovoid in shape (2- 8 µm) , along with few compound starch grains having up to 4 components. phloem were well developed and showed the presence of isolated bast fibres along with usual phloem elements, where phloem parenchyma cells contained starch grains. Wood was wide consisted of vessels, tracheids, fibres and ray. The fibers were heterogenous. Here xylem in the centre were typically composed of thick walled fibre-tracheids (6-8 layers) surrounded by many Vessels equally distributed throughout or occurring singly or in a group of two distributed all over the xylem . Medullary rays were radially elongated and contained starch grains. Vessels were pitted and reticulate.

Root : T.L.S (Fig.121)

Cork cells appeared rectangular. The cells of the cortex also appeared rectangular as of cork , but were larger in size. There were large spindle shaped stone cells in the cortex. Phloem parenchyma were straight. Xylem rays were fairly thick walled, simple pitted and each cell contained 4-6 starch grains.. Tracheids contained 3 to 4 rows of bordered pits. The reticulate thickened vessels were also found.

Root : R.L.S (Fig.122)

The phloem ray cells appeared square to rectangular. They were thin walled and filled with starch grains. The xylem rays also were filled with starch grains. The vessels showed 3-4 rows of bordered pits.

Root : Powder study (Fig.123)

The components present in the powder were thin walled cork, stone cells, ray parenchyma with starch grains, fibre-tracheids, reticulate thickened vessels.

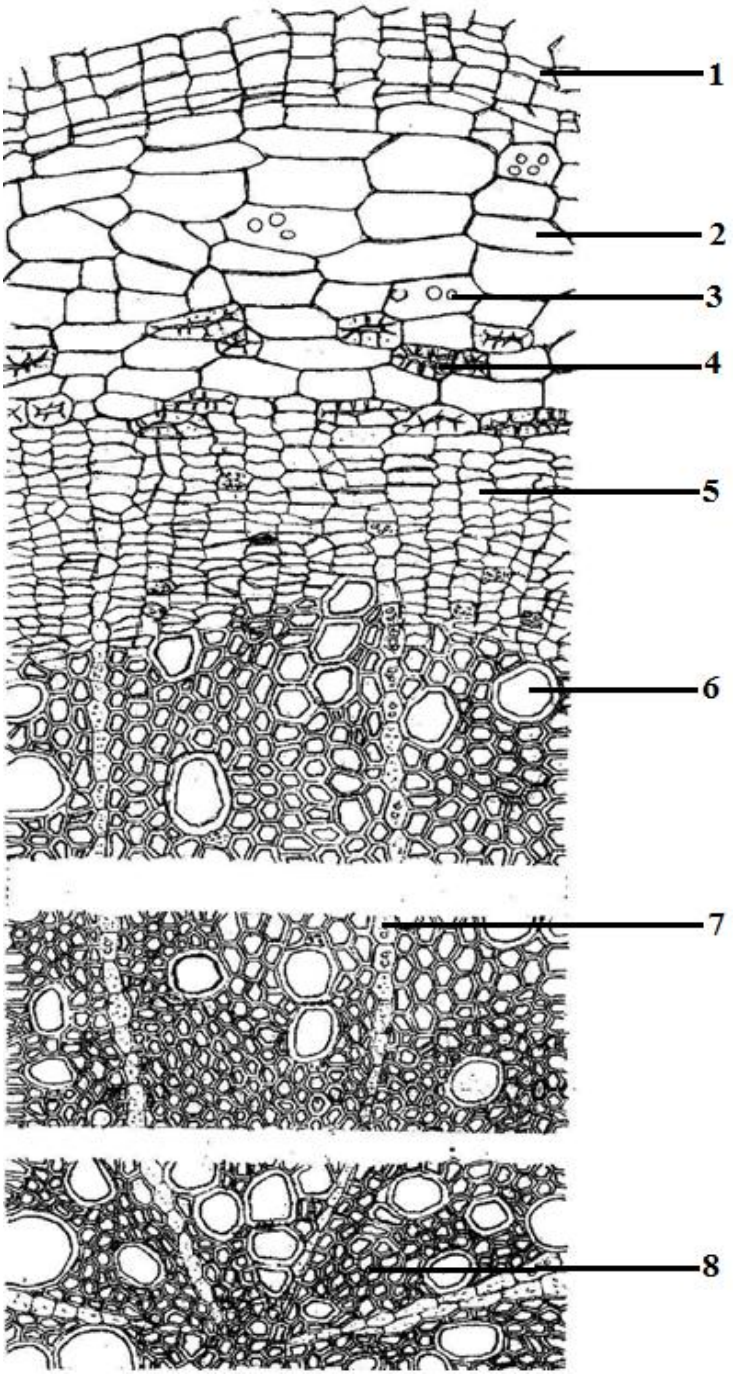


Fig.120. ***Adhatoda vasica*** **root, T.S:** 1. Cork, 2.Cortex, 3.Parenchyma with starch grains, 4.Stone cell, 5.Phloem, 6.Vessels, 7.Xylem rays, 8.Fibre tracheids.

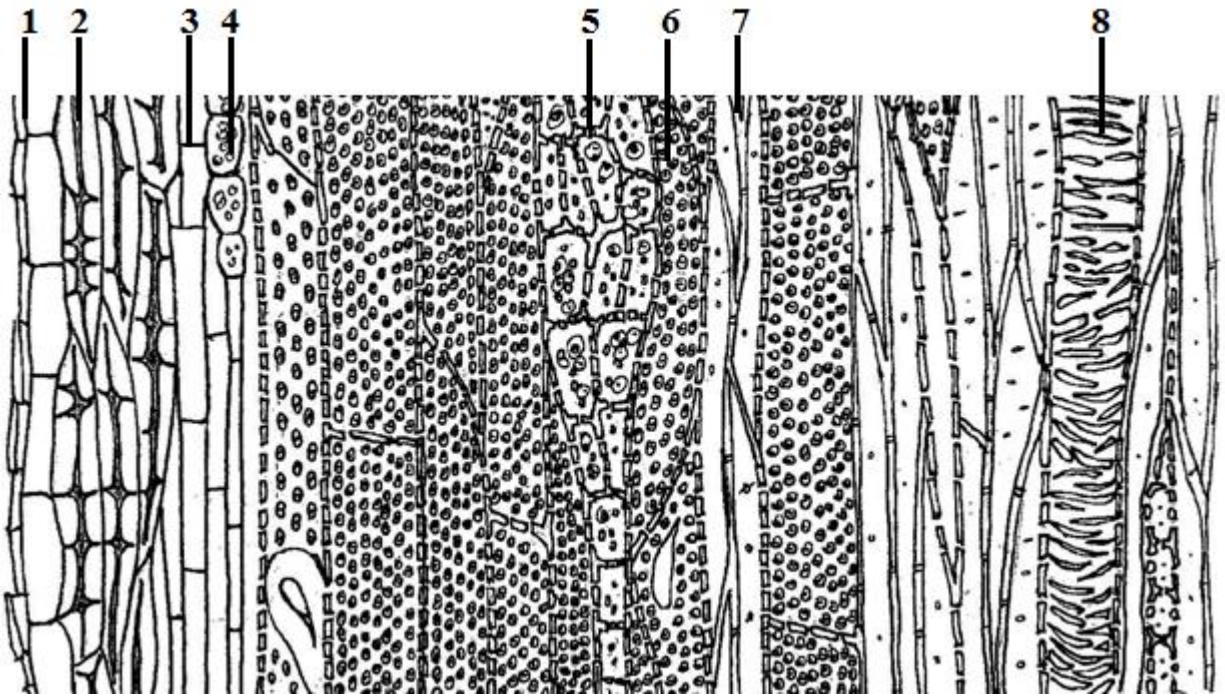


Fig. 121. *Adhatoda vasica* root, T.L.S: 1.Cork, 2.Stone Cell, 3.Phloem parenchyma, 4.Phloem rays, 5.Xylem rays with starch grains, 6.Bordered pitted tracheids, 7.Fibre tracheids, 8.Reticulate thickened vessels.

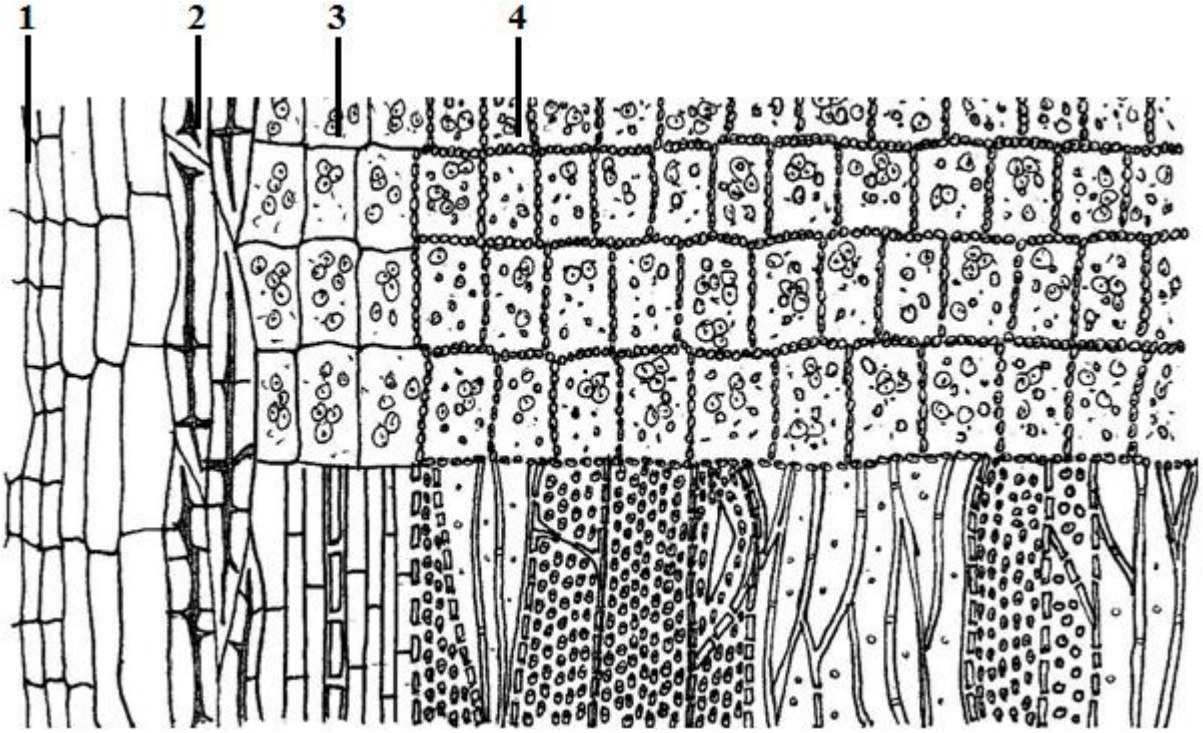


Fig.122. *Adhatoda vasica* root, R.L.S: 1.Cork, 2.Stone Cell, 3. Phloem rays, 4.Xylem rays with starch grains.

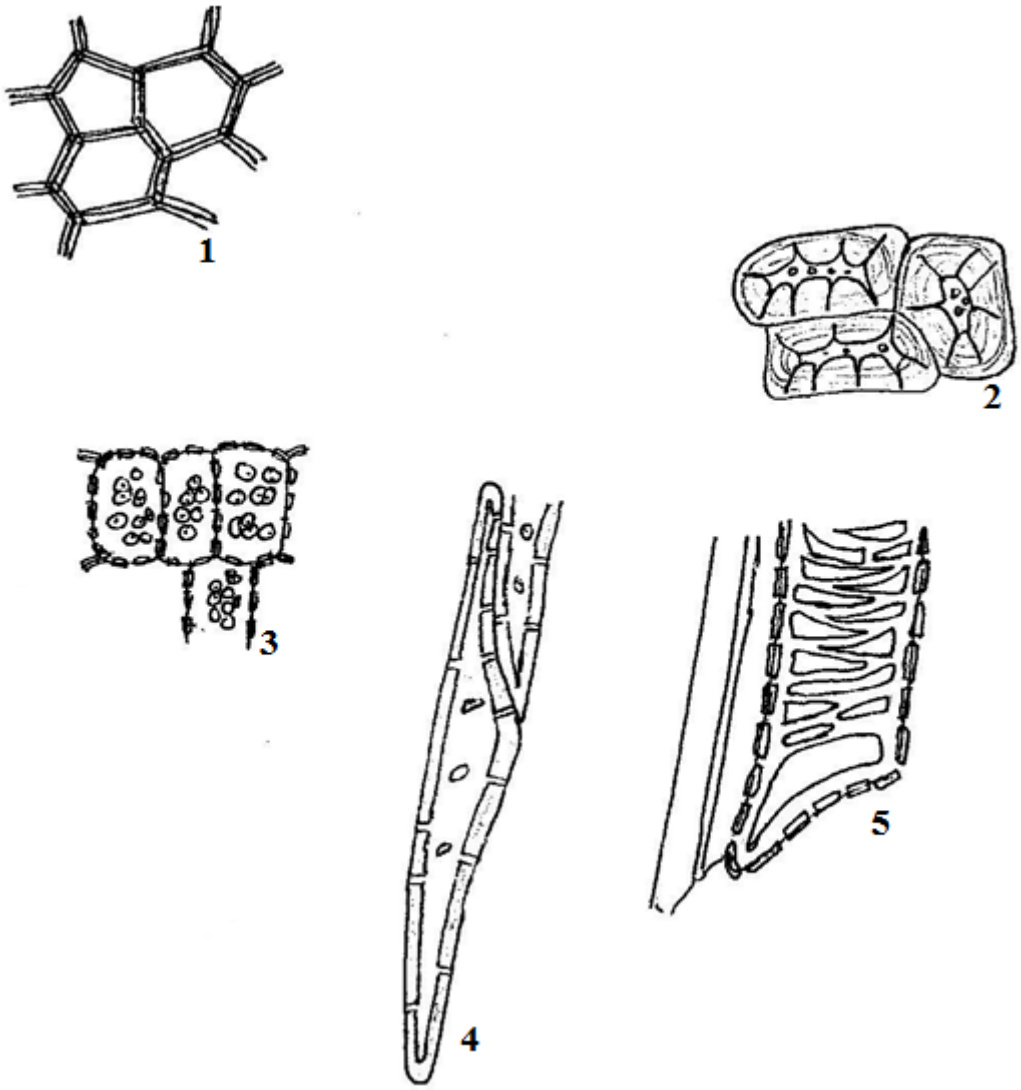


Fig.123. ***Adhatoda vasica*** **root,powder study:**1.Cork, 2.Stone cells, 3.Ray parenchyma with starch grains, 4.Fibre tracheids, 5.Reticulate thickened vessels.

Distinguishing features

Pharmacognostic markers

1. Thin walled cork
2. Starch grains.
3. Stone cells.
4. Xylem in the centre were composed of walled fibre-tracheids.
5. Ray parenchyma containing starch grains.
6. Reticulate thickened vessels.

Phytochemical markers

1. Vanillic acid.
2. Syringic acid.
3. Rhamnose.
4. Xylose.
5. Absence of flavonoids.

Physico-chemical analysis:

Table : Values obtained for the proximate analysis.

Sr.No.	Parameter	Mean ± SD (%)*			Average (%)
		Summer	Monsoon	Winter	
1.	Total Ash Content	04.63±0.31	04.69±0.41	04.66±0.39	4.66
2.	Acid Insoluble Ash content	0.88±0.41	0.89±0.46	0.90±0.39	0.89
3.	Alcohol soluble extractive	06.22±0.16	06.29±0.11	06.13±0.09	6.21
4.	Water soluble extractive	12.11±0.31	12.52±0.13	12.19±0.16	12.27

*Each value is a mean of 3 readings

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6.d. *Polygala chinensis* Linn.(Polygalaceae)

Vernacular names:

English : Indian senega, Common Indian Milkwort.

Gujarat : Pilibhonyasana.

Hindi : Meradu, Miragu.

Marathi : Negli-; Nagpuri : Danaminjo, danaminju, Gurgur.

Distribution and habitat

Throughout India, upto 5,000 ft. (Anon.1956).

Morphological features

An erect branched annual reaching upto 25 cm high, pubescent. Leaves variable, upto 4 cm long, obovate, coriaceous, ciliate, mucronate; petioles hairy. Flowers yellow, in axillary or extra-axillary, short, almost capitate, few-flowered racemes; crest of a single tubular appendage multifid only at the apex; bracts small, membranous. Outer sepals broadly ovate acuminate, with membranous, ciliate margins. Wings herbaceous, oblique, ovate-oblong, acuminate, with narrow, membranous margins ciliate towards the base, longer than the capsule. Capsules orbicular-oblong, strongly ciliate, narrowly margined. Seeds hairy; strophiole glabrous or nearly so, rounded at the apex furnished with 3 membraneous basal appendages.

Medicinal uses:

The root is given in cases of fever and dizziness (Anon.1956).

Previous Phytochemical reports

The plant contained polygalic acid 4.5% and senegin, 2.1% (Hossain *et al.*,1943) and 1, 5–Anhydro-d-mannitol (Alagammal *et al.*,2011).

Previous pharmacognostic reports

No study has been done on the pharmacognostic characters of the root of this plant.

Materials and methods

The plant material has been collected from Timbi village, Vadodara, Gujarat.Phytochemical analysis of roots of the plant for their secondary metabolites such as alkaloids, flavonoids, phenolics, steroids, quinones, mucilages and other compounds were done by standard methods of chromatography and spectrophotometry described in chapter 2. Pharmacognostic, HPTLC finger printing

and physico-chemical studies were done by using standard methods described in chapter 2.

Results

Phytochemistry

The roots of the plant along with the known saponin and steroids were found to contain vanillic, and syringic acids while flavonoids were found to be absent. Mucilage amounted to 7.72 % consisting of rahmnose and xylose.

Pharmacognosy

Macroscopic characters (Fig.124)

The root cylindrical, woody. somewhat tortuous and yellow to yellowish brown colour. Fracture short.

Fig.124. *Polygala chinensis* root.

Microscopic characters

Root : T.S (Fig. 125)

The phellem or cork zone was well developed showed the 4-6 rows of tangentially elongated cells. The walls of these cells were comparatively thick and light brown in colour. Cortex was of 2-5 layers of somewhat broadly rectangular parenchymatous cells, many of them containing yellow amorphous substances. Phloem was made up of usual elements and also was contained yellow amorphous

substances. Phloem rays were found to contain yellowish-brown colouring matter. Secondary xylem was stratified with groups of fiber tracheids along with few isolated pitted parenchyma. Vessels were single and bordered pitted, usually occurred singly. Primary xylem consisted of tracheids having annular thickenings. Xylem rays were uniseriate with few biseriate and contained yellowish-brown colouring matter.

Root : T.L.S (Fig. 126)

The cork cells were many layered and the walls of these cells were comparatively thick and light brown in colour. Phloem rays were thin walled and appeared spindle shaped and some phloem rays contained yellow amorphous substances. Xylem vessels had bordered pits on their walls and few of them showed transversely elongated pits. Primary xylem consisted of tracheids having annular thickenings.

Root : R.L.S. (Fig. 127)

The Phloem and xylem rays consisted of hexagonal shaped pitted cells and contained yellow amorphous substances. Fiber tracheids were thick walled and had simple pits on their walls. Tracheids and vessels were bordered pitted.

Root : Powder study (Fig. 128)

The components present in the powder were cork with brown coloring matter, parenchyma containing yellow amorphous substances, phloem bearing yellow contents, pitted ray parenchyma, wood fibres with broad lumen and tapering ends, fiber tracheids and vessels with annular thickening.

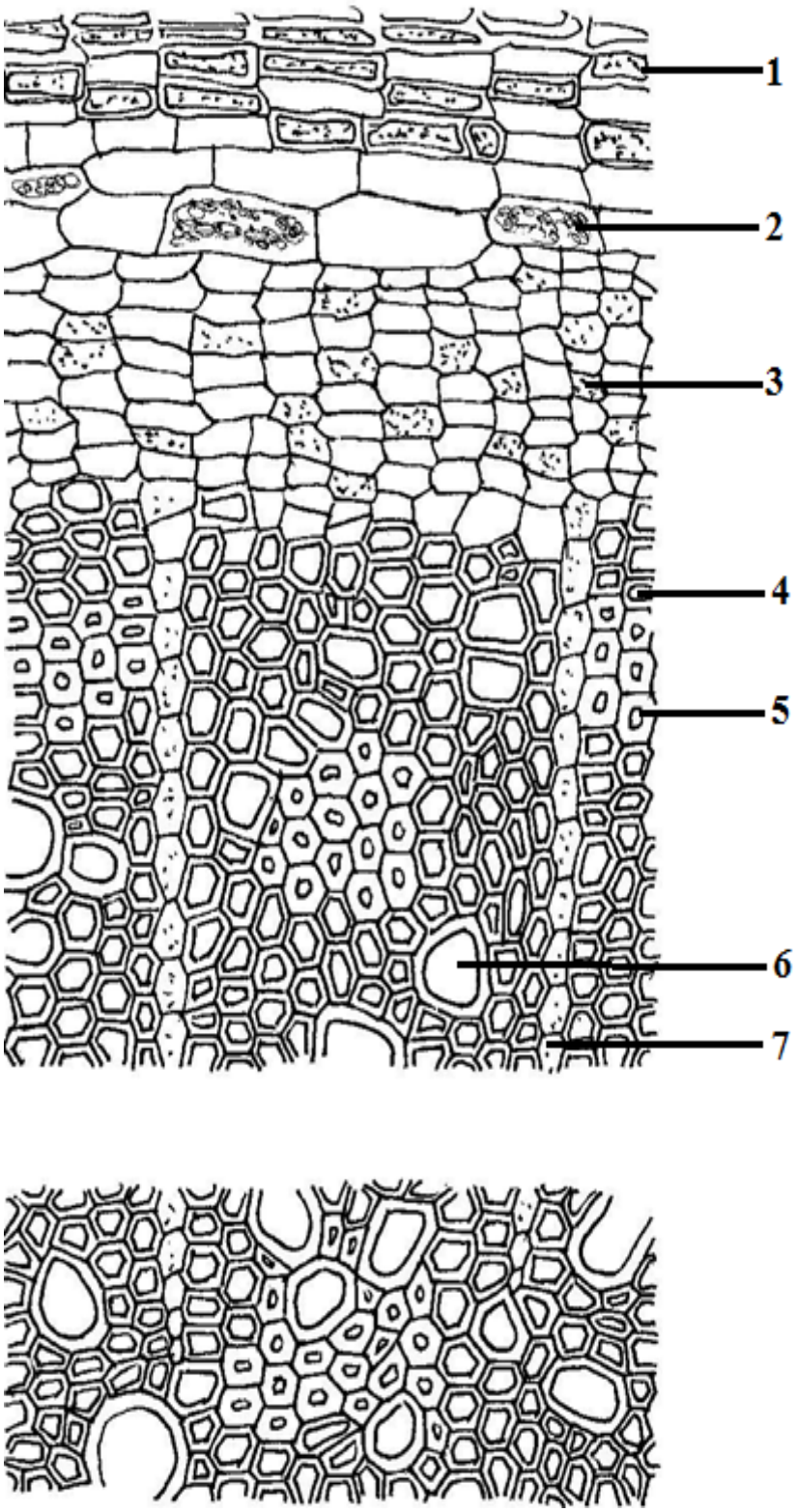


Fig.125.*Polygala chinensis* root, T.S: 1. Cork, 2. Parenchyma with yellow amorphous substances, 3. Phloem with yellow amorphous substances, 4. Xylem, 5.Groups of fibre tracheids, 6. Vessels 7. Xylem rays

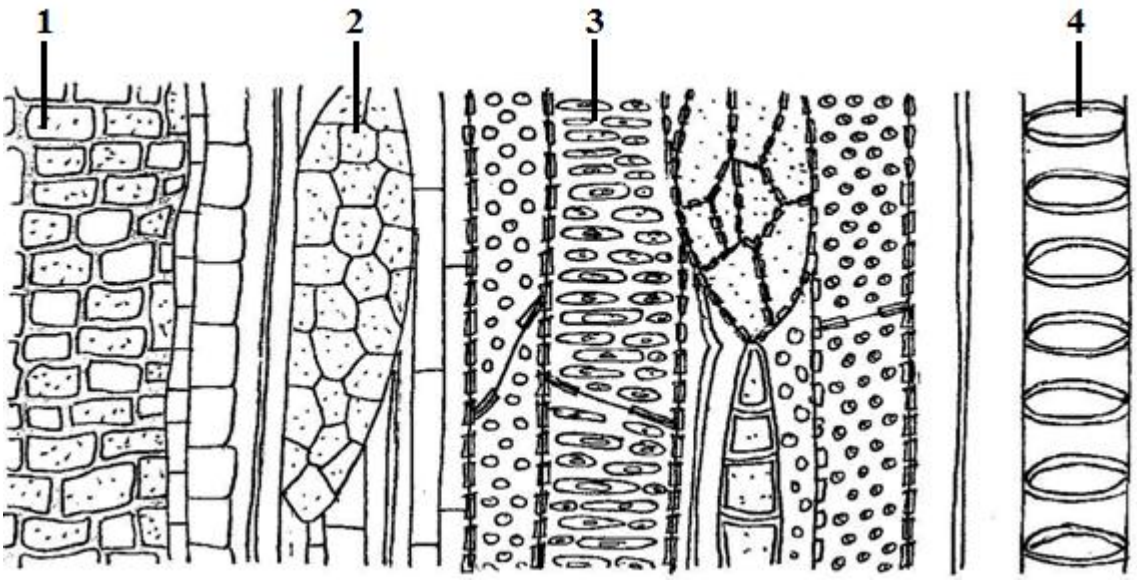


Fig.126. ***Polygala chinensis*** **root, T.L.S**: 1.Cork, 2. Phloem ray, 3.Vessels, 4.Primary xylem.

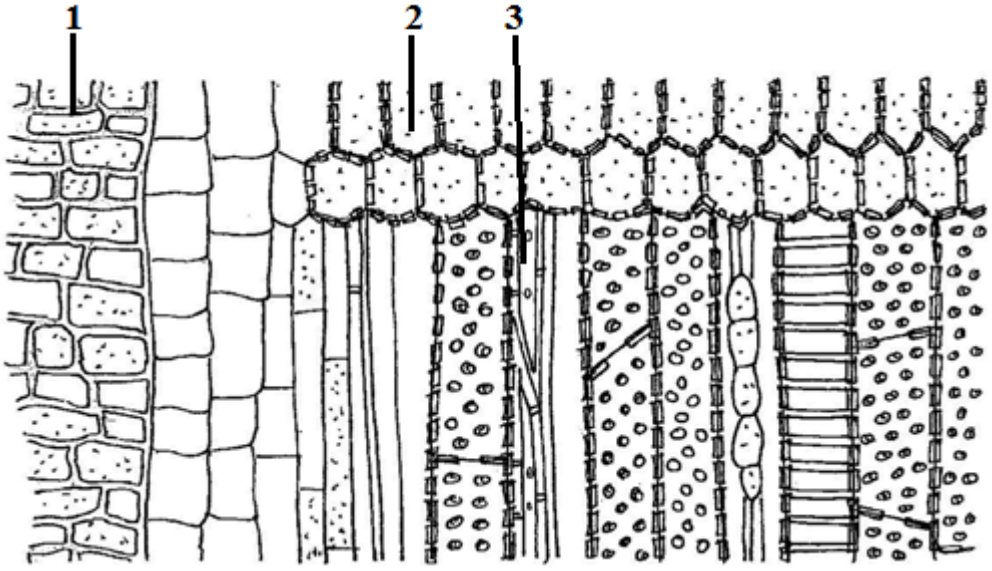


Fig. 127. ***Polygala chinensis*** **root, R.L.S**: 1.Cork, 2. Xylem ray, 3.Fibre tracheids.

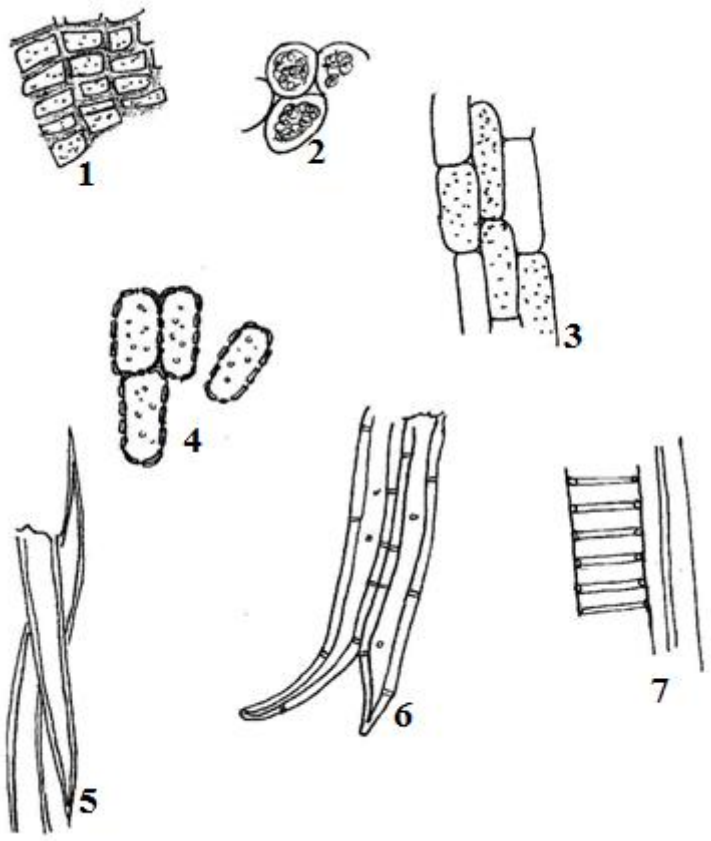


Fig.128.*Polygala chinensis* root,powder study:1.Cork, 2. Parenchyma with yellow amorphous substances, 3. Phloem with yellow amorphous substances,4. Ray parenchyma, 5.Broad lumened fibres, 6.fibre tracheids, 7.Vessels with annular thickening.

Distinguishing features

Phytochemical markers

1. Saponins.
2. Vanillic acid.
3. Syringic acid.
4. Rhamnose
5. Xylose.
6. Absence of flavonoids.

Pharmacognostic markers

1. Cork with brown coloring matter.
2. Parenchyma containing yellow amorphous substances.
3. Fibre tracheids.
4. Vessels with annular thickening.

Physico-chemical analysis:

Table : Values obtained for the proximate analysis.

Sr.No.	Parameter	Mean ± SD (%)*			Average (%)
		Summer	Monsoon	Winter	
1.	Total ash content	4.33±0.42	4.46±0.51	4.32±0.49	4.37
2.	Acid insoluble ash content	0.93±0.11	0.94±0.09	0.93±0.09	0.93
3.	Alcohol soluble extractives	19.22±0.12	19.47±0.26	19.30±0.19	19.33
4.	Water soluble extractives	29.00±0.32	29.81±0.41	29.13±0.19	29.31

*Each value is a mean of 3 readings.

6.e. *Catunaregam spinosa* (Thunb.) Tirveng. (Rubiaceae)

Synonyms: *Xeromphis spinosa* (Thunb.) Keay

Sanskrit : Madana, Pinditak, Dharaphal.

Vernacular names:

Assamese : Gurol, Behmona, Mona.

Bengali : Mainphal.

Hindi : Karhar.

Gujarati : Mindhal, Mindhola, Midhola.

Kanarese : Kare, Banegora, Mangari, Minkare.

Khasia : Diengmakasing-Khlaw.

Kashmir : Kirkla, Kokoa.

Malyalam : Kara.

Marathi : Ghela, Peralu, Mindhal, Wagatta, Gelphal.

Oriya : Palova.

Sanskrit : Madana, Pinditak, Dharaphal.

Tamil : Marukkalankay, Madkarai.

Telugu : Manga.

English : Emetic nut.

Distribution and habitat

Throughout India, common as and undergrowth in the Sal forest ok the Sub-Himalayan tract in many parts of the Indian peninsula.

Morphological features.

A deciduous, thorny shrub or small tree, up to 9m. in height and 90 cm. in girth, bark dark brown grey, tough . Leaves obovate. Flower first white, later turing yellow, fragrant. Berry yellow when ripe, 2.0-3.7 cm. long , globose or broadly ovoid, smooth or obscurely longitudinally ribbed ; seeds many, flat about 4 mm long, angular.

Medicinal uses:

The root is considered cooling, tonic, aphrodisiac, and is used in biliousness and boils of children (Datta and Mukerji,1950). The pulp of the fruit is a valuable emetic. It is nauseant, expectorants, diaphoretic,anthelmintic and abortifacient. It is also useful as a nervine sedative and antispasmodic(Anon.1999).

Previous Phytochemical reports

Fruits yielded ursosaponin, triterpene (m.p. 225-27^{o}), acid resin, yellow essential oil, scopoletin and d-mannitol; triterpene-dirandinin ; dumetoronin A,B,C,D,E,F .Fruit pulp contained sugars, citric and tartaric acids, tannins, pectin and mucilage ; 3-0-(β-D-xylopyranosyloxy) olean-12-en-28-oicacid. Two triterpenic acid sapogenins designated as randialic acid-A and randialic acid-B were reported from the bark while roots contain scopoletin and d-mannitol. Stem heartwood gave α-amyrin, β-sitosterol, oleanolic acid, ursolic acid and D-mannitol. Palmitic, stearic, oleic, linoleic, arachidic and lignoceric acids were present in the seed oil (Anon.1990).

Materials and methods

The plant material has been collected from Rajpipla,Gujarat. Phytochemical analysis of roots of the plant for their secondary metabolites such as alkaloids, flavonoids, phenolics, steroids, quinones, mucilages and other compounds were done by standard methods of chromatography and spectrophotometry described in Chapter 2. Pharmacognostic, HPTLC finger printing and physico-chemical studies were done by using standard methods described in Chapter 2.

Results

Phytochemistry

The root contained high concentration of a coumarin scopoletin along with phenolic acids vanillic and syringic acids while ferulic (*cis*- and *trans*-isomers), melilotic and *p*- coumaric acids were in traces. Mucilage amounted to 6.6% consisting of glucose , xylose and rhamnose . The root also showed the presence of unidentified alkaloids and steroids while saponins were found in good concentrations.

Pharmacognosy

Macroscopic characters (Fig.129)

The root were tap root, cylindrical, woody. The outer surface was light brown in colour and showed longitudinal wrinkles. Fracture fibrous , odour not distinct.

Fig.129. *Catunaregam spinosa* root.

Microscopic characters

Root : T.S (Fig. 130)

The root in T.S was circular in outline. The cork composed of 5-10 layers of thin-walled cells, cells of outer 2 to 4 layers were light brown and rectangular while the cells of inner 3 to 4 layers were more or less cubical in shape and some of them filled with brown content. The secondary cortex was well developed, several layered, consisting of large polygonal thin walled parenchymatous cells most of them filled with starch grains and few with reddish-brown content . There were 3 to 4 continuous layered of stone cells lied in the cortex with few scattered sclerenchymatous fibers and rhomboidal crystals. The starch grains were simple, rounded to oval.Secondary phloem composed of sieve elements and parenchyma , traversed by
phloem rays; some phloem parenchyma found filled with yellowish-brown contents.; phloem rays 1-2 cells wide, isodiametric to slightly radially elongated in inner phloem region and radially elongated in outer phloem region.Wood occupied bulk of root dominated by fibre along with vessels, tracheids and few xylem parenchyma traversed by xylem rays, vessels occured singly or in groups of 2-3 with multiseriate

simple and boarded pits and mostly found associated with fibre tracheids, The fibres were linear with pointed ends, narrow lumen and uniseriate simple pitting found in abundance; xylem parenchyma have simple pits or spiral thickening; xylem rays uni to biseriate, thin-walled, cells redially elongated and pitted filled with starch grains. The starch grains were simple and rounded to oval in shape .

Root : T.L.S (Fig. 131)

The cork cells were compressed and elongated. The cells of the cortex contained starch grains. Narrow lumened stone cells were found associated with rhomboidal crystals.The phloem rays were spindle shaped and contained starch grains. They were mostly biseriate to triseriate. Wood fibres showed simple pits. The vessels showed 3-4 rows of bordered pits.

Root : R.L.S (Fig. 132)

The cork cells appeared flat, compressed and elongated. Cortical cells were polygonal and most of the cells here were filled with starch grains followed by wavy sclerenchymatous fibers. The cortical cells contained reddish-brown content. The phloem and xylem rays were with starch grains. The vessels showed angular pits The primary xylem elements showed spiral thickening.

Root : Powder study (Fig. 133)

The powder is characterized by the presence of brown cork cells, starch grains, groups of stone cells showed both broad and narrow lumen, ray cells with starch grains, wood fibre and vessels with spiral thickening.

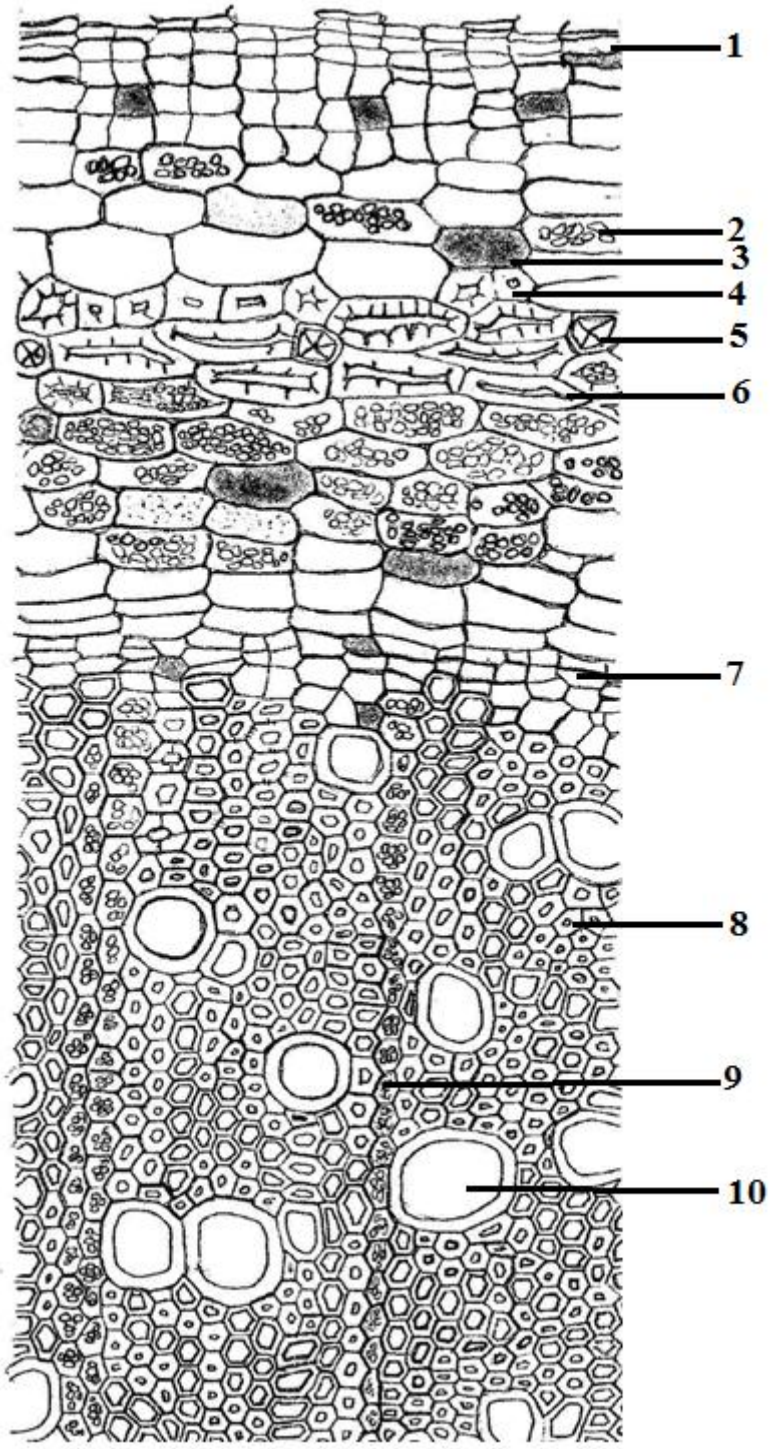


Fig.130. ***Catunaregam spinosa*** **root, T.S :** 1. Cork cell, 2. Starch grains, 3. Reddish-brown content cell, 4. Sclerenchymatous fiber, 5. Stone cell, 6. Rhomboidal crystal, 7.Phloem ray, 8. Wood fibre, 9. Xylem rays. 10.Vessel.

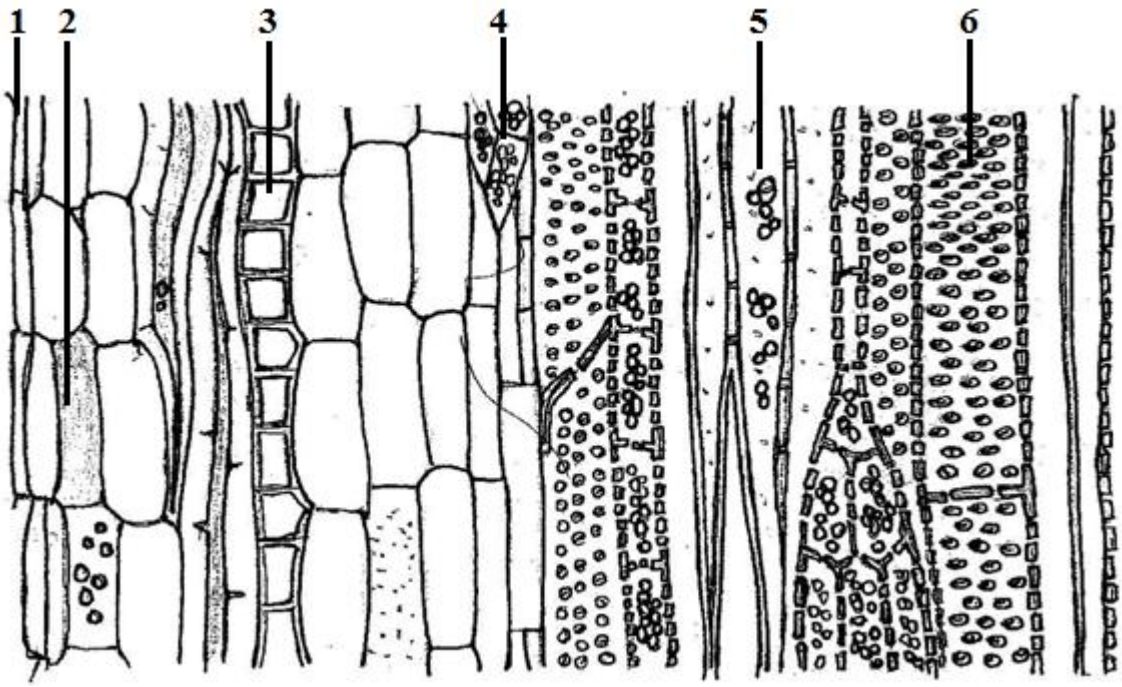


Fig. 131. ***Catunaregam spinosa*** **root, T.L.S:** 1. Cork cells, 2. large polygonal Parenchyma, 3. Redish-brown content cell, 4. Rhomboidal crystal, 5. Phloem ray, 6. Wood fibre, 7.Vessels.

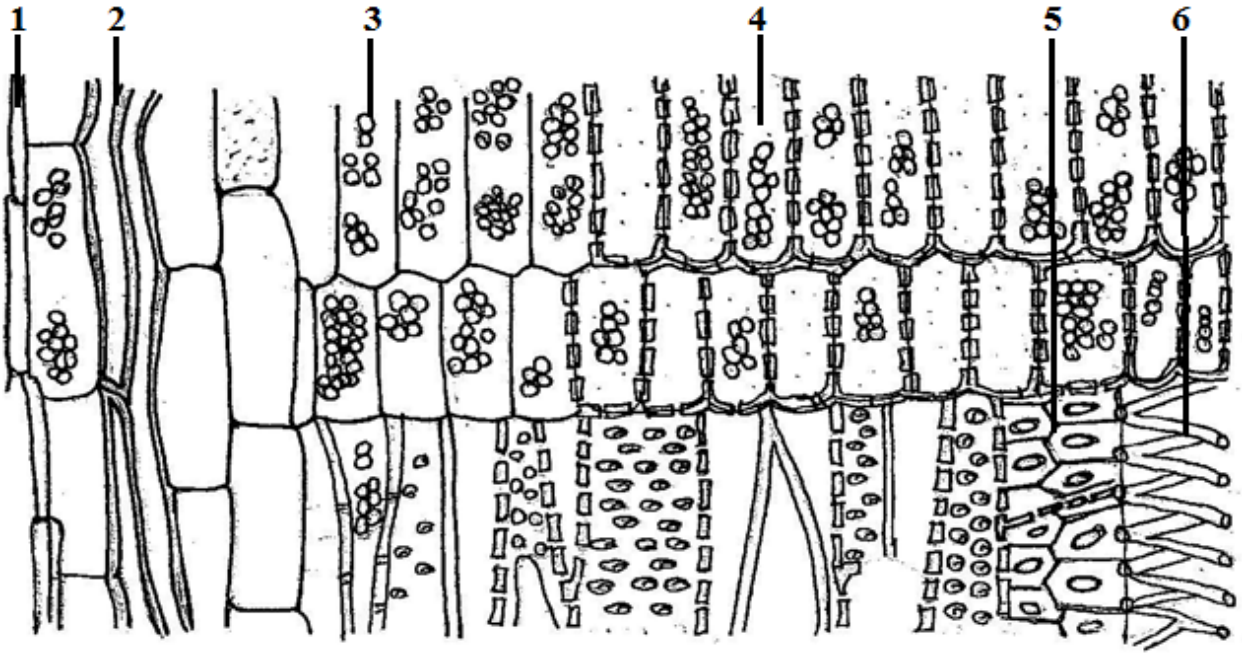


Fig.132.*Catunaregam spinosa* root,R.L.S:1.Cork cells, 2.Sclerenchymatous fiber, 3. Phloem rays with starch grains, 4. Xylem rays, 5.Angular pitted vessel, 6.Spiral vessels.

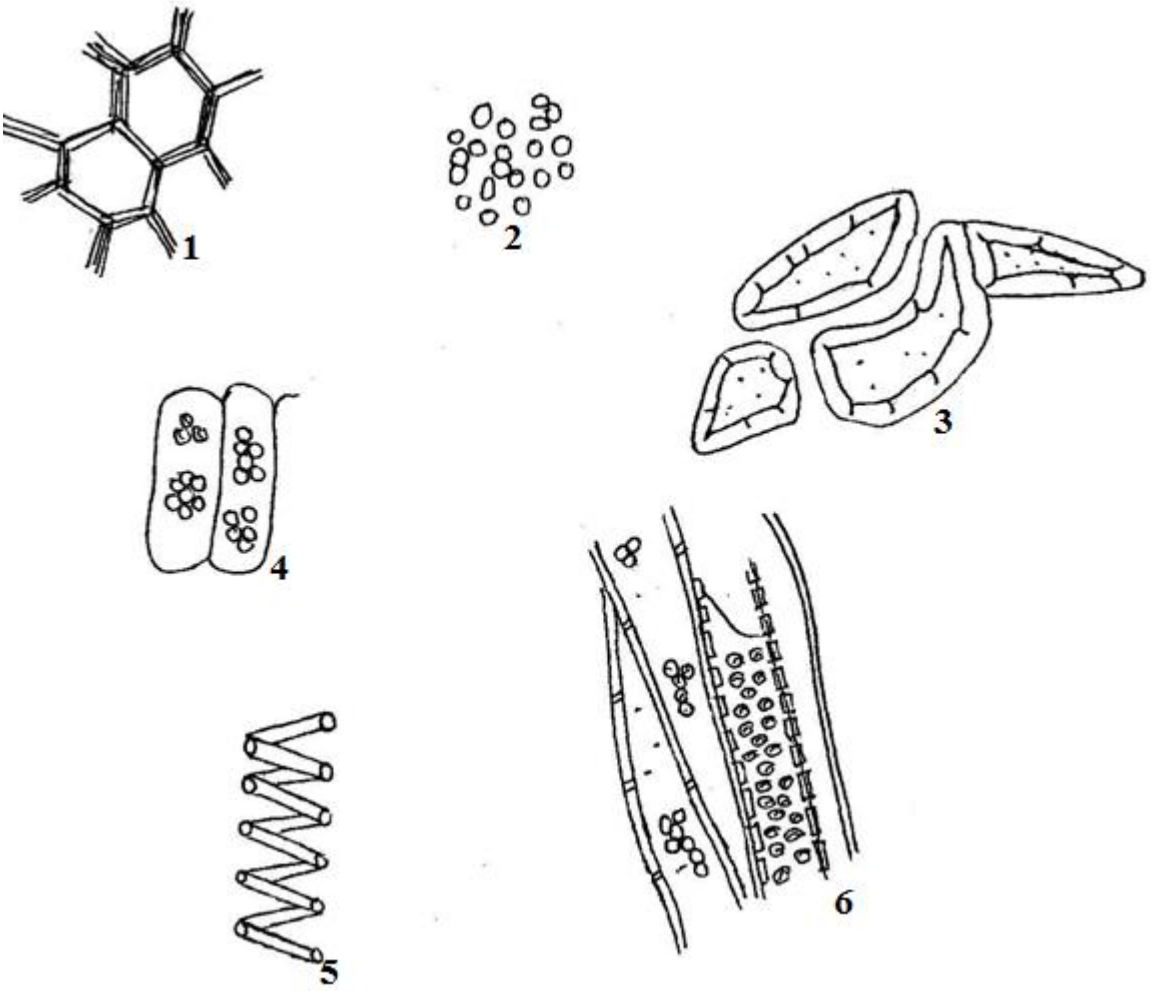


Fig.133. ***Catunaregam spinosa*** **root, Powder study:**1.Brown cork cells, 2.Starch grains, 3.Stone cells, 4.Ray cells with starch grains, 5. Spiral thickened vessel, 6.Wood fibres associated with vessels.

Distinguishing features

Pharmacognostic markers

1. Brown cork cells.
2. Parenchyma containing reddish-brown contents.
3. Starch grains.
4. Stone cells.
5. Sclerenchymatous fibers.
6. Reticulate thickened vessels.

Phytochemical markers

1. Scopoletin.
2. Vanillic acid.
3. Syringic acid.
4. Rhamnose.
5. Xylose.

Physico-chemical analysis:

Table : Values obtained for the proximate analysis.

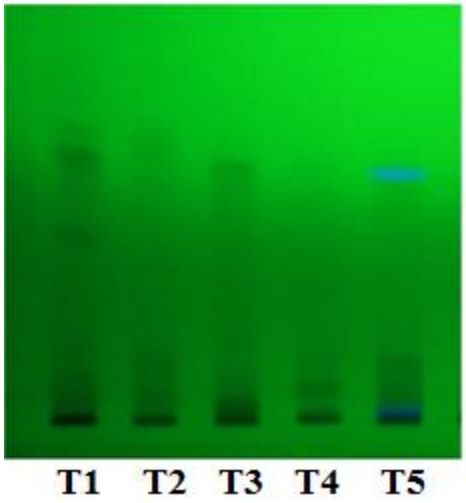
Sr.No.	Parameter	Mean ± SD (%)*			Average (%)
		Summer	Monsoon	Winter	
1.	Total Ash Content	03.13±0.21	03.24±0.32	03.16±0.22	3.18
2.	Acid Insoluble Ash content	0.68±0.41	0.71±0.33	0.68±0.46	0.69
3.	Alcohol soluble extractive	08.62±0.19	08.69±0.23	08.23±0.16	8.51
4.	Water soluble extractive	14.01±0.20	15.02±0.19	14.13±0.13	14.39

*Each value is a mean of 3 readings

6.f. HPTLC fingerprinting and Physo-chemical analysis of *Polygala senega* and its substitutes/adulterants

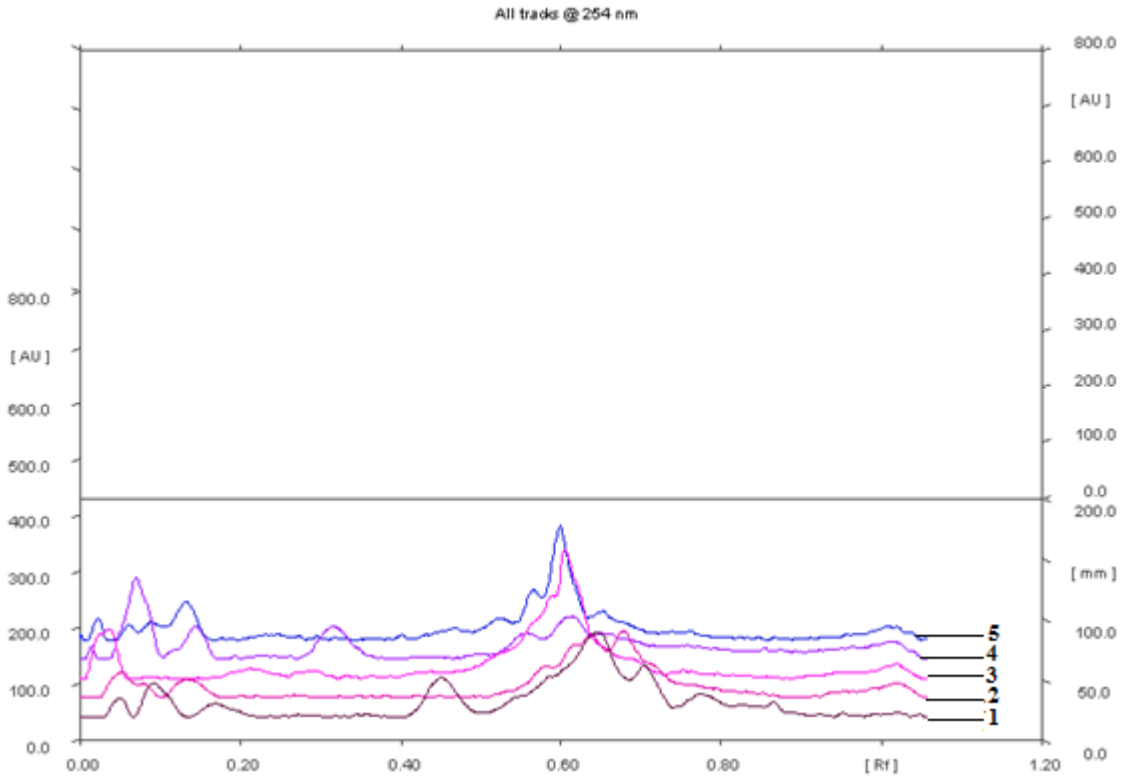
HPTLC fingerprinting

Figure 133.a : HPTLC chromatogram of *Polygala senega* and its substitutes/adulterants. (UV 254 nm).



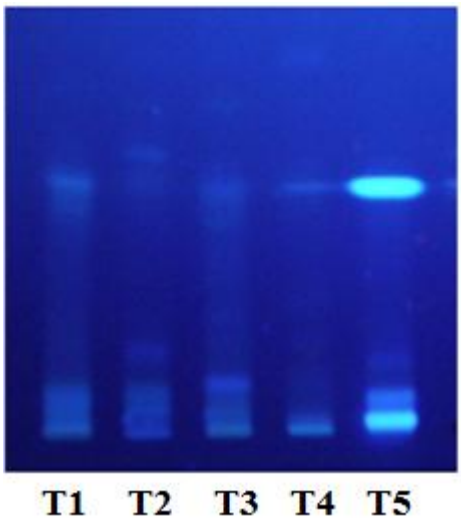
(a).T1-*Polygala senega,* T2-*Polygala chinensis,*T3-*Acalypha indica,*T4-*Adhatoda vasica,* T5-*Catunaregam spinosa* .

Figure 133.b : HPTLC chromatogram of *Polygala senega* and its substitutes/adulterants. (UV 254 nm).



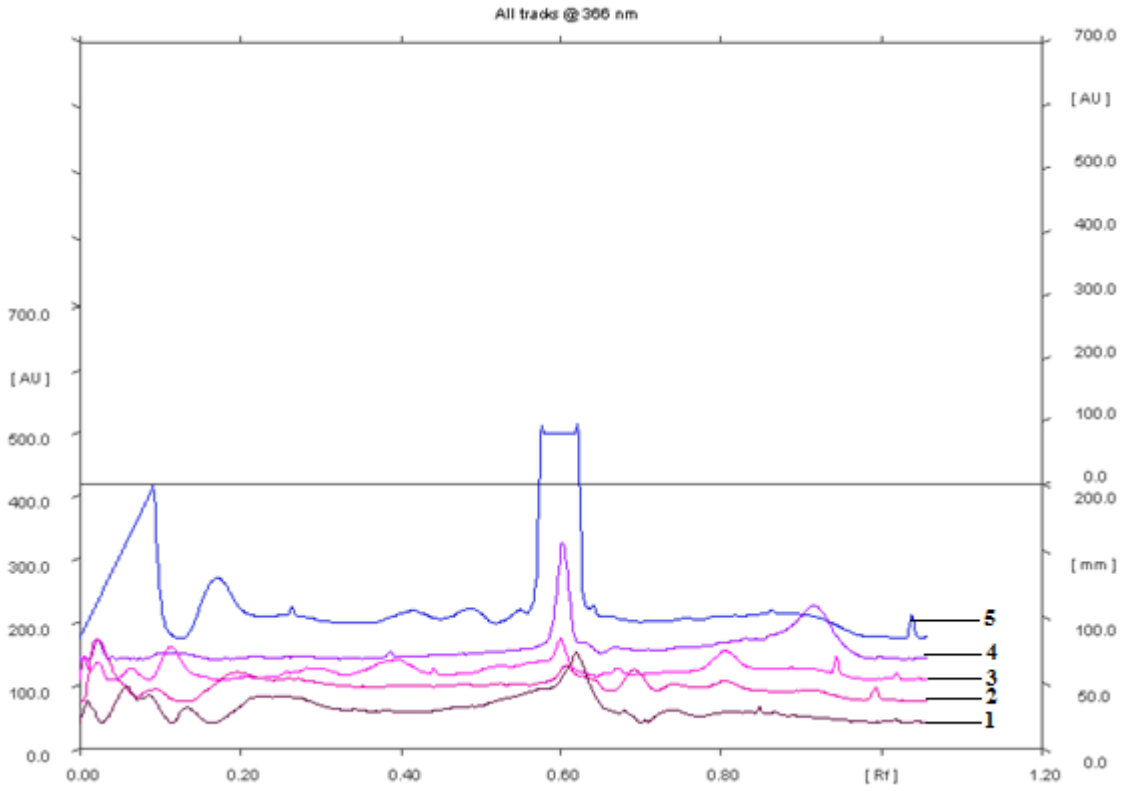
(b).1-*Polygala senega*, 2-*Polygala chinensis*,3-*Acalypha indica*,4-*Adhatoda vasica*, 5-*Catunaregam spinosa* .

Figure 133.c: HPTLC chromatogram of *Polygala senega* and its substitutes/adulterants (UV 366 nm).



(a).T1-*Polygala senega,* T2-*Polygala chinensis,*T3-*Acalypha indica,*T4-*Adhatoda vasica,* T5-*Catunaregam spinosa* .

Figure 133.d : HPTLC chromatogram of *Polygala senega* and its substitutes/adulterants (UV 366 nm).



(b).1-*Polygala senega,* 2-*Polygala chinensis,*3-*Acalypha indica,*4-*Adhatoda vasica,* 5-*Catunaregam spinosa* .

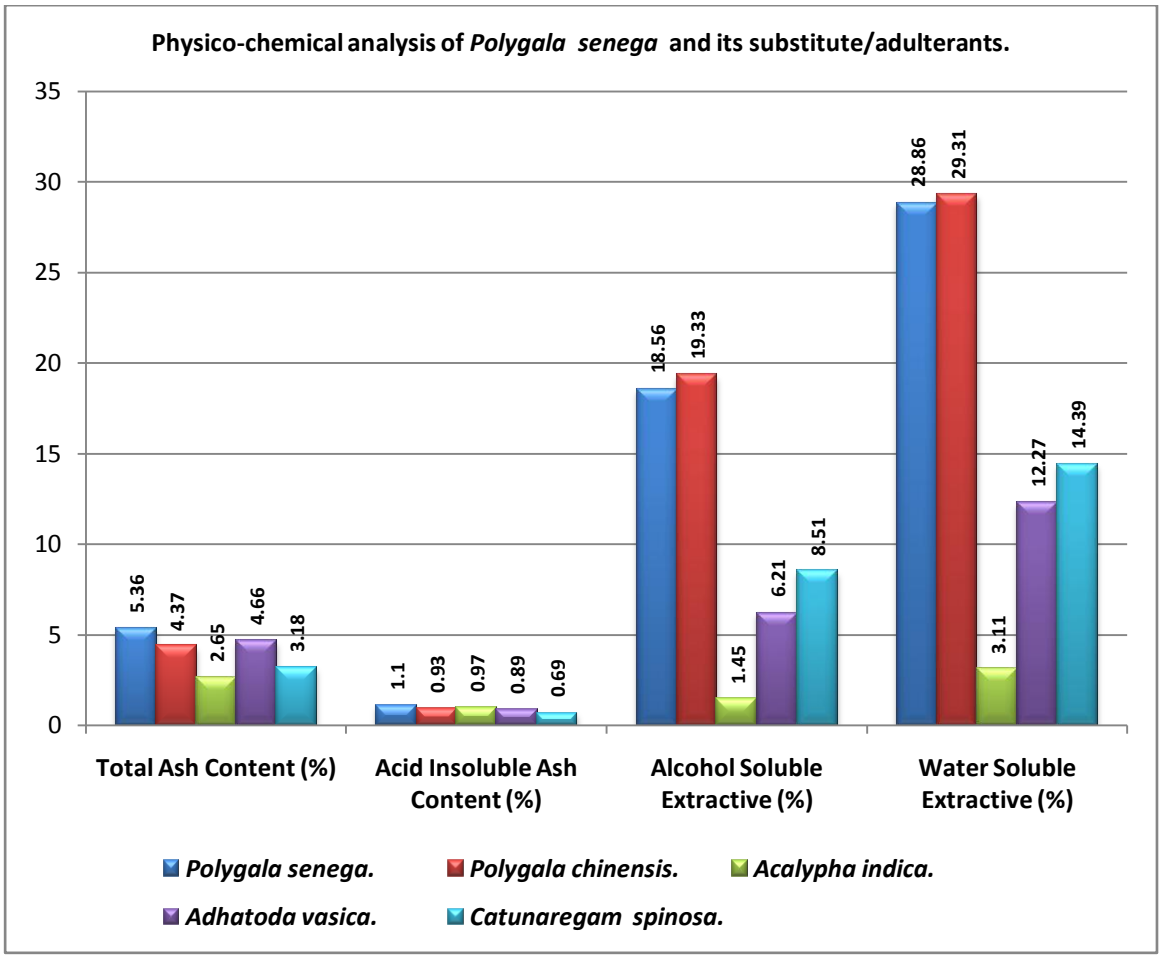
HPTLC profile of *Polygala senega* showed the presence of 8 peaks when observed under UV 254 nm (figure-133.b)and 10 peaks in 366 nm (figure-133.d). There were 3 major peaks found at R_f 0.45, R_f 0.65 and R_f 0.70 under UV 254 and 4 peaks at R_f 0.23, R_f 0.26 , R_f 0.57, and R_f 0.62 under 366 nm. The *Polygala chinensis* and *Adhatoda vasica* showed the presence of 8 peaks , *Acalypha indica* 6 peaks and *Catunaregam spinosa* 11 peaks when observed under UV 254 nm.Under UV366 nm, *Polygala chinensis* and *Catunaregam spinosa* both were showed the presence of 10 peaks while *Acalypha indica* showed the presence of 11 peaks and *Adhatoda vasica* 5 peaks.

HPTLC profile of *Polygala senega* and its substitutes/adulterants observed under UV 254 nm (figure-133.b) showed that *Catunaregam spinosa* was similar in 2 peaks but differed in 9 peaks. Both *Polygala chinensis* and *Adhatoda vasica* were similar in 1 peak but differed in 7 peaks, while *Acalypha indica* was not show any peak similar to that of *Polygala senega* but differed in having 6 peaks.

HPTLC profile of *Polygala senega* and its substitutes/adulterants observed under UV 366 nm (figure-133.d) showed that *Catunaregam spinosa* was similar in 4 peaks but differed in 6 peaks. *Polygala chinensis* was similar in 2 peaks and differed in 8 peaks. *Acalypha indica* was similar in 1 peak and differ in 10 peaks while *Adhatoda vasica* did not show any peak similar to that of *Polygala senega* but differed in 5 peaks.

Physico-chemical analysis

Physico-chemical analysis of *Polygala senega* and its substitutes/adulterants.



Total ash content

Total Ash Content of *Polygala senega*(5.39 %) along the material collected in different season does not show significant variation (Table-) while the closest value to the substitute/adulterant in descending order is 4.66 % (*Adhatoda vasica*), 4.37% (*Polygala chinensis*), 3.18 % (*Catunaregam spinosa*), and 2.65 % (*Acalypha indica*).

Acid insoluble ash content

Acid insoluble ash content of *Polygala senega*(1.10 %) along the material collected in different season does not show significant variation (Table-) while the closest value to the substitute/adulterant in descending order is *Acalypha indica* (0.97 %), *Polygala chinensis* (0.93 %), *Adhatoda vasica* (0.89 %) and *Catunaregam spinosa* (0.69%).

Amongst the substitutes/adulterants of *Polygala senega* the *Polygala chinensis* showed the closest value of total ash content which showed that the *P. chinensis* was more close to *P.senega* as compared to other substitutes/adulterants of *P. senega*.

Alcohol soluble extractive

Alcohol soluble extractive value of *Polygala senega* (18.56%) along the material collected in different season does not show significant variation (Table-) while the closest while the closest value to the substitute/adulterant was of *Polygala chinensis* (19.33%) which also showed the maximum extraction. The values of *Catunaregam spinosa Adhatoda vasica* and *Acalypha indica* was found to be 8.51 %, 6.21% and 1.45 % respectively.

Water soluble extractive

Water soluble extractive value of *Polygala senega* (28.86 %) along the material collected in different season does not show significant variation (Table-) while the closest value to the substitute/adulterant was of *Polygala chinensis* (29.31%) which also showed the maximum extraction. The values of *Catunaregam spinosa Adhatoda vasica* and *Acalypha indica* was found to be 14.39 %, 12.27% and 3.11 % respectively.

Amongst the substitutes/adulterants of *Polygala senega*, the *Polygala chinensis* showed the maximum extraction of phytoconstituents and the extractive values were also very close to *P. senega* while the extractive values of other substitutes/adulterants were less than the half values of *P. senega* which reflect that the *P. Chinensis* could be better substitute as compared to other substitutes/adulterants.

Chapter 7

7.a. *Saraca indica* Linn. (Caesalpiniaceae)

Synonyms: ***Saraca asoca*** (Roxb.) De Wilde , *Jonesia asoca* Roxb.

Sanskrit : Anganapriya, Apashoka, Ashoka, Vitashoka

Vernacular names:.

Assamese : Ashoka.

Bengali : Ashoka.

English : Asok Tree.

Gujrati : Ashoka.

Hindi : Ashoka.

Kannada : Ashokadamara, Ashokamara, Kankalimara.

Kashmiri : Ashok.

Malayalam : Asokam.

Marathi : Ashok.

Oriya : Ashoka.

Punjabi : Asok.

Tamil : Asogam, Asogu, Asokam.

Telugu : Asogam, Asokamu, Vanjulamu ,Ashokapatta.

Distribution and habitat

The plant is a small to medium sized, evergreen tree distributed throughout India, particularly in Central and Eastern Himalayas, ascending to 2000 ft.

Morphological features

The plant has numerous spreading somewhat drooping branches bearing nearly sessile large a bruptly pinnate leaves, one to two feet long, having two to three pairs of large oblong lanceolate leaflets, large dense corymbs of brilliant orange-red fragrant flowers, and rigidly coriaceous or almost woody smooth turgid pods about six inches long containing four to eight seeds.

Medicinal uses:

The bark is bitter, astringent, sweet, refrigent, anthelmintic, styptic, stomachic, constipating, demulcent. It has stimu sating effect on endometrium and the ovarian tissue. It is useful in dyspepsia, fever, biliousness, burning sensation, abnormal enlargement of visceral organs, colic, dysentery, internal bleeding, haemorrhoids,

ulcers, uterine affections, menorrhagia especially due to uterine fibroids, meno-metrorrhagia, leucorrhoea and pimples. Leaves possess blood purifying properties and its juice mixed with cumin seed is used to cure gastralgia. Flowers are considered as excellent uyerine tonic and are used in cervical adenitis, biliousness, syphilis, hyperdipsia, burning sensation, haemorrhagic dysentery, piles, scabies in children and inflammation. Dried flowers are used in diabetes. Seeds are used in treating bone fracturyes, strangury and vesical calculi(Anon. 2005).

Previous Phytochemical reports

The bark of plant presence of (-) epicatechin, procyanidin, β- 2,11'-deoxypro**c**yanidin B, (+) catechin, (24, £)- 24- methyl-cholesta-5-en-3p-ol, (22 E, 21£)-24-ethycholesta-5,22 dien-33-ol, (24 £)-24- ethylcholesta-5-en-3-β-ol,leucopelargonidin-3-O-β-Dglucoside,leucopelargonidin and leucocyanidin. The flower part of plant contains oleic, linoleic, palmitic and stearic acids, β-sitosterol, quercetin, kaempferol- 3-0-β-D- glucoside, quercetin- 3-0-β-D-glucoside, apigenin- 7-0-β-D-glucoside, pelargonidin- 3, 5- diglucoside, cyanidin-3, 5-diglucoside, palmitic, stearic, linolenic, linoleic, β and γ sitosterols, leucocyanidin and gallic acid. Seed and Pod contains oleic, linoleic, palmitic and stearic acids, catechol,(-)epicatechol and leucocyanidin . Five lignan glycosides, lyoniside, nudiposide, 5-methoxy-9-β-xylopyranosyl-(−)-isolariciresinol, icariside E3, and schizandriside, and three flavonoids, (−)-epicatechin, epiafzelechin-(4β→8)-epicatechin and procyanidin B2, together with β-sitosterol glucoside, were isolated from dried bark (Pradhan, 2009).

Previous pharmacognostic reports

Little data is available on pharmacognosy of the stem bark of this plant. (Anon.1990 and Malati G and Pillai 2005).

Materials and methods

The plant material has been collected from Vadodara, Gujarat. Phytochemical analysis of roots of the plant for their secondary metabolites such as alkaloids, flavonoids, phenolics, steroids, quinones, mucilages and other compounds were done by standard methods of chromatography and spectrophotometry described in Chapter 2. Pharmacognostic, HPTLC finger printing and physico-chemical studies were done by using standard methods described in Chapter 2.

Results

Phytochemistry

The stem bark is found to contain cyanidin. The phenolic acids were vanillic and syringic acids. Mucilage amounted to 3.2% consisting of rhamnose, glucose and xylose. The plant also showed the presence of unidentified flavonoids and steroids.

Pharmacognosy

Macroscopic characters (Fig.134.)

Stem bark was channelled , externally rough, grayish brown and showed warty protuberances and numerous prominent circular to transversely elongated lenticels with transverse and longitudinal cracks. The fracture was short and slightly fibrous.

Fig.134. *Saraca indica* bark.

Microscopic characters

Bark : T.S (Fig. 135)

The T.S. of stem bark showed the periderm composed of 6 to 16 rows of slightly tangentially elongatedcork cells. The cells had brown thick walls. The outer few rows of cork cells were much compressed and their cell walls were wavy with outer one or two rows ruptured due to the formation of rhytidoma. Some of these cells contain reddish brown contents. The Phellogen was single row of narrow tangentially elongated thin walled cells. The phelloderm, innermost two to three rows of the cork made up of polygonal to isodiametric shaped cells and the walls were thin and yellow, interspersed in which were few thick walled, narrow lumened stone cells, which mostly isodiametric in shape with few cubical to linear shaped. The middle bark which was mainly secondary cortex was composed of thin walled

parenchymatous cells. The cells were fairly large polygonal and compactly arranged with few stone cells. Small spherical starch grains were also occurred in most of the parenchyma cells, while others contain rhomboidal crystals of calcium oxalate of various sizes and yellow masses. Inner to the secondary cortex was one or two nearly continuous tangential bands of stone cells. The stone cells were thick walled, with simple pits, striations and narrow lumened. The inner bark which constitutes nearly half the thickness of the entire bark consisted of phloem tissue, bast fibres and medullary rays. The phloem parenchyma cells were small thin walled, many of them contained yellow amorphous contents, spherical starch grains and rhomboidal crystals. Alternating with the parenohymatous elements were also found small groups of fibres (each group consisting of 4 to 6 cells). The fibers were of both septate and aseptate and many of these fibers were found associated with rhomboidal crystals. There were few compressed and nearly obliterated thin walled phloem elements with a light yellow colour occured in between the regular tissues. The medullary rays were mostly uni-or bi-seriate and most of them contained rhomboidal crystals. The cells were radially elongate at outer and appeared tangentially elongated in the inner.

Bark : Powder study (Fig. 136)

The components present in the powder were thick walled brown colour cork cells, parenchyma containing spherical starch grains, stone cells with thick walled, sclerides, rhomboidal crystals, crystal fibres.

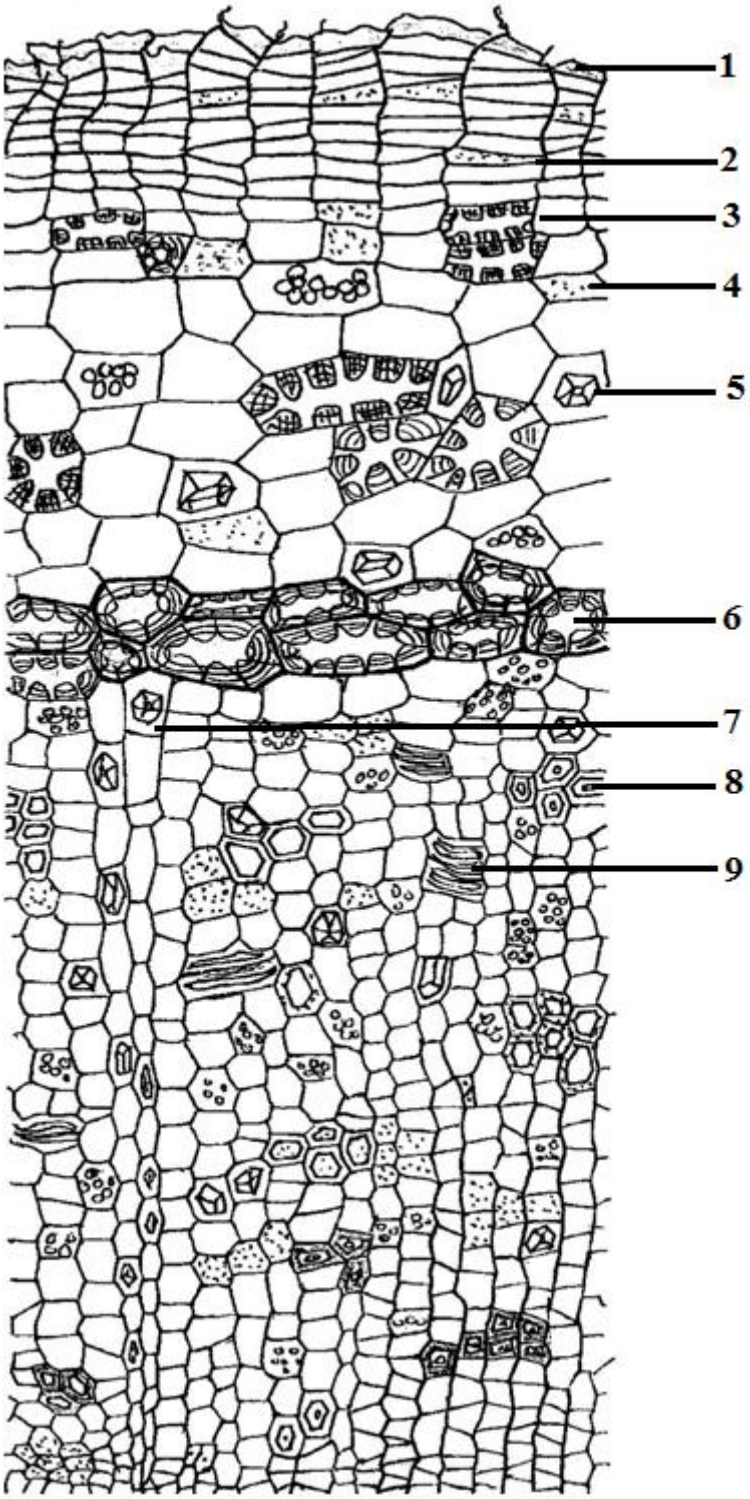


Fig. 135. *Saraca indica* bark, T.S: 1.Cork, 2.Phellogen, 3. Phelloderm with stone cell, 4. Parenchyma with yellow masses, 5. Rhomboidal crystal, 6. Stone cell, 7. Phloem rays, 8. Phloem fibres group, 8.Compressed obliterated thin walled phloem elements.

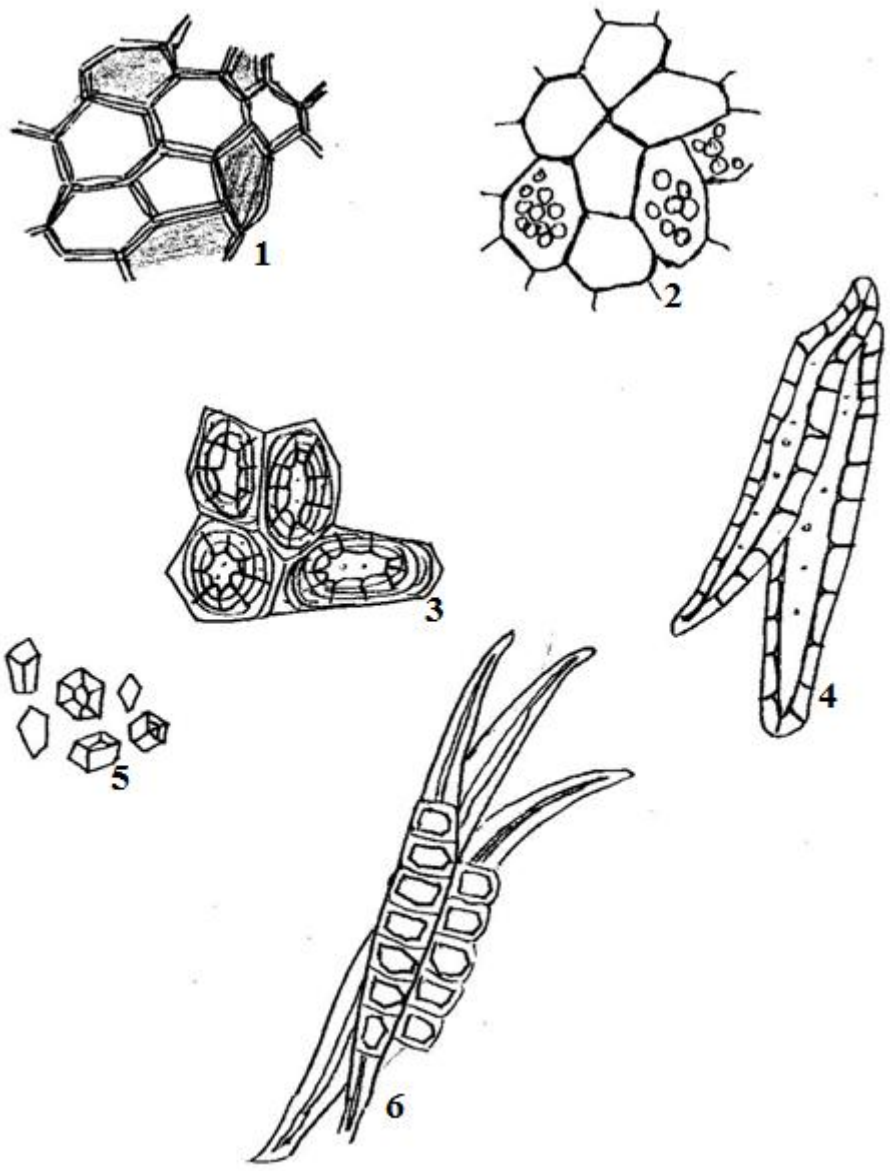


Fig. 136. *Saraca indica* bark, powder study: 1.Thick walled brown colour cork cells, 2.Parenchyma containing spherical starch grains, 3.Stone cells, 4.Sclerides, 5.Rhomboidal crystals, 6.Crystal fibres.

Distinguishing features

Phytochemical markers

1. Cyanidin.
2. Vanillic acid.
3. Syringic acid.
4. Glucose.
5. Xylose.

Pharmacognostic markers

1. Thick walled brown colour cork cells.
2. Parenchyma containing spherical starch grains.
3. Thick walled narrow lumened stone cells.
4. Sclereids.
5. Rhomboidal crystals.
6. fibers were septet and aseptet.
7. Crystal fibres.

Physico-chemical analysis:

Table : Values obtained for the proximate analysis.

Sr.No.	Parameter	Mean ± SD (%)*			Average (%)
		Summer	Monsoon	Winter	
1.	Total Ash Content	10..28±0.14	10.26±0.19	10.18±0.22	10.24
2.	Acid Insoluble Ash content	0.99±0.46	1.03±0.31	0.99±0.24	1.00
3.	Alcohol soluble extractive	18.76±0.16	18.09±0.09	18.13±0.19	18.33
4.	Water soluble extractive	14.26±0.33	14.10±0.27	14.08±0.21	14.15

*Each value is a mean of 3 readings.

7.b.*Bauhinia variegata* Linn. (Caesalpiniaceae)

Sanskrit : Kancanaraka, Gandari.

Vernacular names

Assamese : Kotora, Kurol.

Bengali : Raktakanchan.

English : Orchid Tree.

Hindi : Goriyal, Barial, Gurial, Gwiar, Kachnar, Papri.

Kannada : Arisinantige, Ayata, Bilikanjivala, Irkubalitu, Kondaalka, Kovindaara.

Malayalam : Chovanna-Mandaru, Kovidaram, Chuvannamundiri, Unnu.

Manipuri : Chingthao-Angouba.

Marathi : Kavidara, Kanchan, , Raktakanchan, Thaur.

Mizoram : Vau-Favang, Vaube, Vaufawang.

Nepali : Takki.

Oriya : Vau-Favang, Vaube, Kachan.

Tamil : Mandarai, Segappumandarai.

Telugu : Bodanta, Daevakanchanamu, Kaanchanamu, Mandaara.

Distribution and habitat

The plant is a medium sized deciduous tree found wild in the sub-Himalayan tract and outer Himalaya upto 1300 m.,in Punjab, dry forests of Eastern, Central and South India, Assam, Sikkim, Chota Nagpur, Western Peninsula. Also cultivated largely as a garden and roadside.

Morphological features

The plant is an erect branched tree 7-10 m high. Leaves 10-12 cms. in diameter, roundish, about as broad as long, divided 1/3- ½ the way down into 2 obtuse or subacute lobes, faintly puberulous beneath, base cordate; main nerves 9-11; petioles upto 3 cm long, glabrous; stipules triangular-oblong, acute, pubescent, deciduous. Flowers in terminal and axillary few-flowered corymbosa racemes; bracts beneath the pedicels triangular, acute, pubescent; pedicels pubescent; 2-bracteolate below the middle. Calyx-tube slightly dilated upwards; limb upto 2 cm long, splitting into 2 coriaceous segments slightly divided at the

apex into 5 short teeth. Petals 5 upto 5 cms. long, subequal, erect, imbricate, the upper inner, oblanceolate, acute, with a long claw, white, rose or purple. Fertile stamens 3-4. Ovary with long stalk; ovules 16-20; style long; stigma oblique, peltate. Pods upto 30 cms., flat. Seeds 12-16, oblong-ellipsoid.

Medicinal uses

The bark of this plant is traditionally used for tonic, astrain; ulcers.it is also useful in skin disease (Manandhar, 2002). The bark is alterative, anthelmintic, astringent and tonic. The juice of the bark is used in the treatment of amoebic dysentery, diarrhoea and other stomach disorders. A paste of the bark is useful in the treatment of cuts and wounds, skin diseases, scrofula and ulcers. It can also be used in cough conditions, asthma, abdominal distention, also act as a gargle for sore throats, prevent from skin diseases, or internally as a remedy for diarrhea. It is helpful in managing skin discoloration (Gordon and David 2001, Vileges *et.al.*,1997).

Previous Phytochemical reports

The stem bark showed presence of hentriacontane, octacosanol, stigmasterol. (Prakash and Khosa 1976) and of sterols, glycosides, reducing sugars and nitrogenous substances . (Prakash, and Khosa 1978).The stem yielded a flavonone glycoside characterized as 5, 7-dihydroxyflavonone-4 –O – Z -L – rhanmopyranosyl-e – D – glucopyranoside (Gupta *et.al.*,1979).The isolation of e-sitosterol, lupeol, kaempferol-3-glucoside and a 5, 7-dimethoxyflavonone-4 –O –Z – L – rhanmopyranosyl-e- D-glucopyranoside was also reported from the stem of the plant.(Gupta *et.al.*,1980, Duret and Paris 1977). Kaempferol-3-glucoside, was isolated from stem of this plant(Gupta and Chauhan. 1984). A new phenanthraquinone, named bauhinione, has also been isolated and its structure has been elucidated as 2, 7-dimethoxy-3-methyl-9, 10-dihydrophenanthrene-1, 4-dione on the basis of spectroscopic analysis (Zhao *et.al*., 2005).

Previous pharmacognostic reports

Only data on T.S. was available on pharmacognosy of the stem bark of this plant. (Anon.1990, Anon.1999 and Malati and Pillai 2005).

Materials and methods

The plant material has been collected from Rajpipala, Gujarat. Phytochemical analysis of roots of the plant for their secondary metabolites such as alkaloids, flavonoids, phenolics, steroids, quinones, mucilages and other compounds were done by standard methods of chromatography and spectrophotometry described in Chapter 2. Pharmacognostic, HPTLC finger printing and physico-chemical studies were done by using standard methods described in Chapter 2.

Results

Phytochemistry

The stem bark is found to contain flavonoe kaempferol. The phenolic acids were vanillic syringic, protocatechuic and o-coumaric acids. Mucilage amounted to 4.3% consisting of rhamnose and glucose. The plant also showed the presence of unidentified and steroids.

Pharmacognosy

Macroscopic characters (Fig.137)

Stem bark was curved , externally rough, grayish brown and showed exfoliations and small transverse and longitudinal cracks and fissures with prominent longitudinal ridges. The fracture was outer short and inner fibrous.

Fig. 137. *Bauhinia variegata* bark

Microscopic characters

Bark : T.S (Fig. 138)

The T.S. of stem bark showed the cork made up of 12 to 16 rows of rectangular cells. The outer 3 to 5 rows of cells were slightly compressed and had thin light brown walls, While inner 3 to 9 rows of cells were broad rectangular with thick brown walls. Due to the formation of rhytidoma outer one or two rows of cork were found ruptured. The Phellogen was single row of narrow tangentially elongated thin walled cells. The phelloderm consisted of broad rectangular yellowish brown cells. They were 3 to 5 layered thick and showed the presence of isolated stone cells and pericyclic fibers in it. The secondary cortex was made up of tangentially elongated to isodiametric cells, the cells were thin-walled parenchyma. Some of these cells contained spherical starch grains, reddish-orange contents and abundant crystals. The crystals were of rosette, prismatic and rhomboidal types. Many small groups of stone cells and pericyclic fibres were also found scattered in this region. The stone cells were mostly elongated and thin walled, broad lumened with prominent pits and striations. The pericyclic fibres were broad, thick-walled with narrow lumened. The inner bark consisted of phloem tissue, bast fibres and medullary rays. This region was dominated by fibres, found in small groups of 6 to 12 fibres in each group, many of them found associated with rosette, prismatic or rhomboidal crystals . The phloem parenchyma cells were small thin walled, many of them contained rosette crystals and few spherical starch grains. Stone cells either isolated or associated with fibres were distributed throughout the phloem region. The stone cells were characteristically found associated with crystal fibers. The phloem fibers were thick-walled and narrow lumened. The medullary rays were mostly uni-or bi-seriate and funnel shaped. The cells were tangentially elongated in outer region and radially elongated in inner region and many of them containing rosette crystals. The stone cells were also present in the medullary rays.

Bark : Powder study (Fig. 139.)

The components present in the powder were thin and thick walled cork cells, parenchyma containing rosette crystals, spherical starch grains, prismatic and rhomboidal crystals, stone cells with thin walled and broad lumen, sclereids, phloem

ray containing rosette crystals, stone cells associated with crystal fibres, thick walled and narrow lumened fibers.

Distinguishing features

Phytochemical markers

1. Kaempferol.
2. *O*-Coumaric acid.
3. Vanillic acid.
4. Syringic acid.
5. Glucose.
6. Rhamnose.

Pharmacognostic markers

1. Thin and thick walled cork cells.
2. Parenchyma containing reddish-orange contents.
3. Starch grains.
4. Association of stone cells with crystal fibers.
5. Sclereids.
6. Rhomboidal and prismatic crystals.
7. Crystal fibres.
8. Stone cells present in the medullary rays.

Physico-chemical analysis:

Table : Values obtained for the proximate analysis.

Sr.No.	Parameter	Mean ± SD (%)*			Average (%)
		Summer	Monsoon	Winter	
1.	Total Ash Content	7.88±0.42	7.72±0.16	7.93±0.39	7.84
2.	Acid Insoluble Ash content	3.13±0.05	3.05±0.03	3.20±0.06	3.13
3.	Alcohol soluble extractive	11.23±0.63	12.02±0.60	11.67±0.77	11.64
4.	Water soluble extractive	8.36±0.42	8.98±0.25	8.40±0.33	8.58

*Each value is a mean of 3 reading

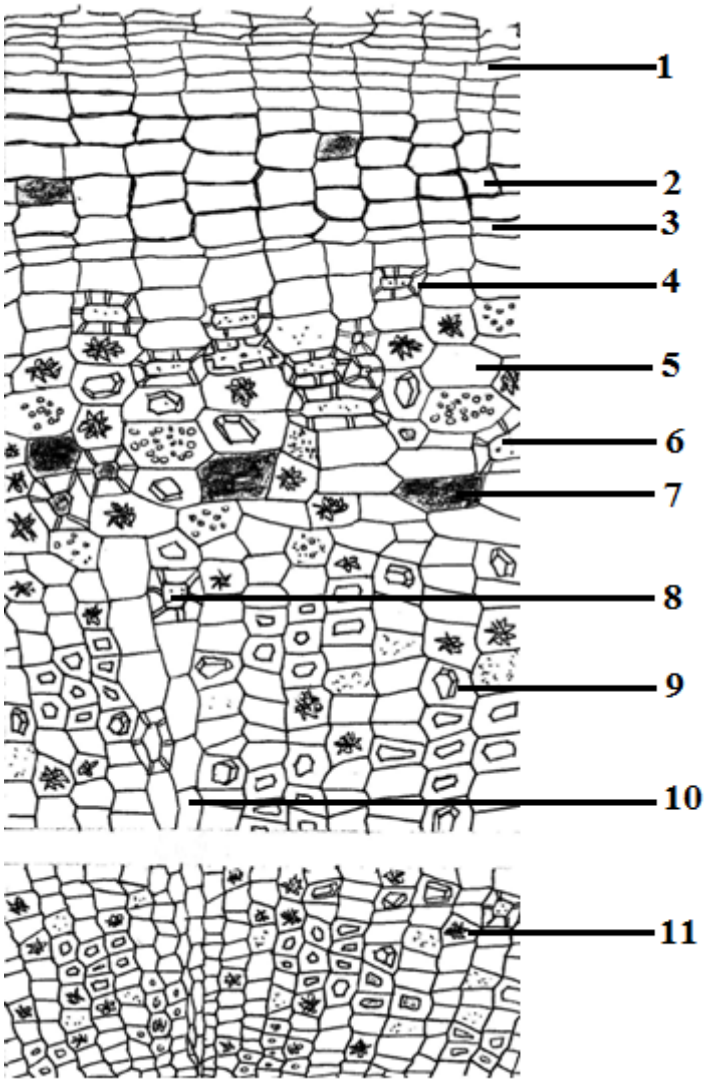


Fig. 138. ***Bauhinia variegata*** **bark, T.S**: 1.Thin walled cork cells, 2. Thick walled cork cells, 3. Phellogen, , 4. Phelloderm with stone cell, 5. Secondary cortex, 6.Sclereids, 7. Parenchyma containing reddish-orange contents, 8. Stone cells, 9.Rhomboidal crystals, 10.Phloem rays, 11. Rosette crystals.

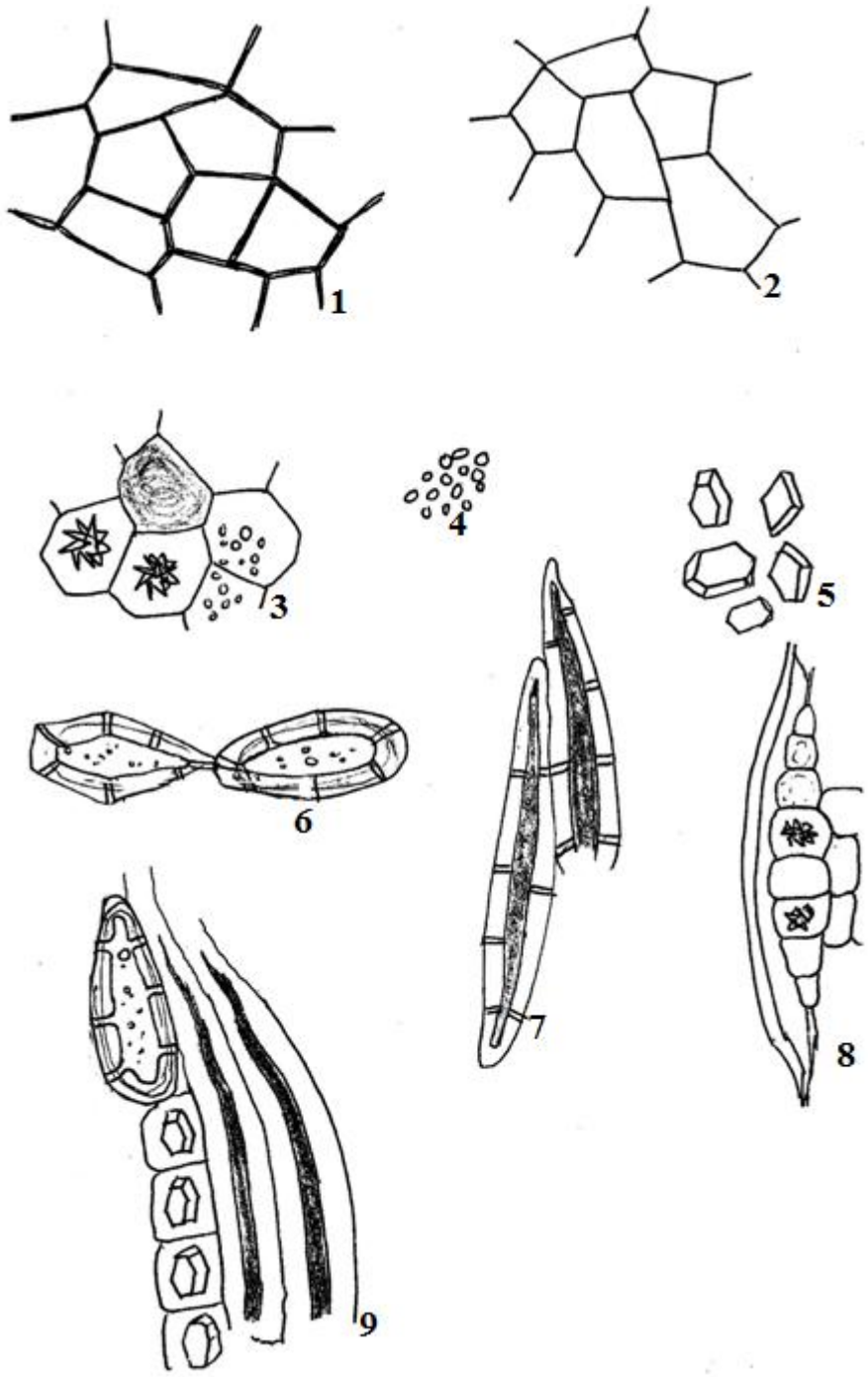


Fig. 139. ***Bauhinia variegata*** **bark, powder study**: 1.Thick walled cork cells, 2.Thin walled cork cells, 3.Parenchyma containing reddish-orange contents and rosette crystals , 4. Spherical starch grains, 5.Rhomboidal and prismatic crystals, 6.Stone cells, 7.Sclereids, 8.Phloem rays with rosette crystals, 9.Stone cells associated with crystal fibers.

7.c. *Bombax ceiba* Linn.(Bombacaceae)

Synonyms: *Bombax malabaricum* DC. , *Salmalia malabarica* (DC.) Schott &Endl.

Sanskrit: Salmali.

Vernacular names:

Assamese : Dumboil, Simalu.

Bengali : Simul, Shimool.

English : Silk Cotton Tree, Indian Silk Cotton Tree, Simul.

Hindi : Kantisembal, Pagun, Ragatsemal, Ragatsembal, Semal, Semul, Semur.

Kannada : Apurani, Buraga, Burla, Dudi, Elava, Hatti, Kempuburaga, Mullelava.

Malayalam: Ilavu, Mocha, Mullilavu, Poola, Semul.

Manipuri :Tera.

Marathi : Kanta-Sair, Savar, Saur, Semal,Simlo, Tamari, Vhadli-Savar.

Mizoram : Pang, Phunchawng.

Oriya : Pang, Phunchawng, Similikonta.

Tamil : Agigi, Ilavam, Ilavu, Kongu, Mullilavu, Pongar, Pulai, Purami, Sallagi.

Telugu : Buraga, Kondaburaga, Mundlaburaga, Pinnaburaga, Salmali.

Distribution and habitat

The plant is deciduous tree distributed throughout the hotter parts of the country upto 1500 m or more.

Morphological features

A tall tree with branches covered by small hard conical prickles. Leaves palmately compound with a long petiole; stipules triangular, caducous. Leaflets 5-7, glabrous, entire, elliptic-lanceolate, acuminate, attenuate at base, more or less leathery, unequal. Inflorescence many fascicles of 1-4 flowers borne, at or near the end of branches. Flowers large, showy, red (occasionally yellow or white); pedicel thick. Calyx 3-lobed (rarely 2-lobed), cup-shaped, smooth outside, densely silky within. Petals twisted in bud, stellate tomentose outside, sparcely pubescent inside, elliptic-oblong, usually recurved. Stamens c. 75 polyadelphous, united at base in 6 phalanges, each of 11-15 stamens, the inner-most phalange surrounding the pistil is composed of 15 stamens of which 5-innermost are the largest and forked; filaments flattened at base; anthers long, afterward twisted, violet. Ovary conical, green, covered with silky hairs; style simple 5.9-6.5 cm long; stigmas 5, filiform. Capsule

10-12.5 cm long; oblong, woody, 5 valved, profusely to finely tomentose. Seeds brown, smooth, obovid, 6 mm long, embeded in silky white wool.

Medicinal uses:

The roots are sweet, cooling, stimulant, tonic and demulcent, and are used in dysentery. The gum is astringent, cooling. stimulant, aphrodisiac, tonic, styptic and demulcent. It is useful in dysentery, haemoptysis of pulmonary tuberculosis, influenza, menorrhagia, burning sensation, strangury, haemorrhoids, blood impurities and vitiated conditions of pitta. The bark is mucilaginous, demulcent and emetic, and is used for fomenting and healing wounds. A paste of it is good for skin eruptions. Leaves are good for strangury and skin eruptions. Flowers are astringent and are good for skin troubles,.splenomegaly and haemorrhoids. Young fruits are useful in calculus affections, chronic inflammations and ulceration of the bladder and kidney. Seeds are useful in treating gonorrhoea, chronic cystitis (Warrier *et.al*.,1994).

Previous Phytochemical reports

The flowers showed presence of β-D-glucoside of β-sitosterol, free β-sitosterol, hentriacontane, hentriacontanol, kaempferol, quercetin and traces of an essential oil (Gopal and Gupta, 1972). The fresh petals of flowers were reported to yield two anthocyanidin glycosides named A and B which were characterized as pelargonidin-5β-D-glucopyranoside and cyanidin-7-methyl-ether-3β-glucopyranoside, respectively (Niranjan and Gupta. 1973). The seeds contained n-hexacosanol, palmitic acid, octabecyl palmitate, gallic acid, tannic acid, l-gallayl-β-glucose, ethyl gallate and a mixture of α-β- and γ-tocopherols (Dhar and Munjal, 1976). Riboflavin and thiamine were reported from the gum (Broker and Bhat. 1953).The stem bark was reported to contain lupeol and β-sitosterol (Mukherjee and Roy, 1971). In a preliminary study, the stem bark showed absence of saponins, alkaloids and flavonoids (Kapoor *et. al*., 1969).The root afforded n-triacontanol, β-sitosterol and glycoside, identified as 5,7,3',4'- tetrahydroxy -6-methoxyflavan-3-0-β-D-glucopyranosyl-α-D-xylopyranoside (Chauhan *et. al*., 1980).

Previous pharmacognostic reports

Only data on T.S. was available on pharmacognosy of the stem bark of this plant. (Anon.2001, and Malati G and Pillai 2005).

Materials and methods

The plant material has been collected from Vadodara, Gujarat. Phytochemical analysis of roots of the plant for their secondary metabolites such as alkaloids,

flavonoids, phenolics, steroids, quinones, mucilages and other compounds were done by standard methods of chromatography and spectrophotometry described in Chapter 2. Pharmacognostic, HPTLC finger printing and physico-chemical studies were done by using standard methods described in Chapter 2.

Results

Phytochemistry

The stem bark is found to contain cyanidin. The phenolic acids were vanillic, syringic and ferulic *(cis*- and *trans*- isomers) acids. Mucilage amounted to 4.3% consisting of rhamnose, galactose, arabinose and glucose. The plant also showed the presence of unidentified steroids.

Pharmacognosy

Macroscopic characters (Fig.140.)

Stem bark was slightly curved , externally gray in colour, rough with fragments of prickles and transverse and longitudinal cracks and. The fracture was fibrous.

Fig.140. *Bombax ceiba* bark.

Microscopic characters

Bark : T.S (Fig. 141.)

The T.S. of stem bark showed the cork made up of 6 to 18 rows of rectangular thin walled stratified cells. The outer 3 to 6 rows of cells were slightly compressed

and had brown walls, while inner 3 to 10 rows of cells were broad rectangular. Due to the formation of rhytidoma outer one or two rows of cork were found ruptured. The Phellogen was indistinct. The phelloderm consisted of broad rectangular and many of them contained orange brown contents. The stone cells were distributed in throughout cork were singly or in paired. The secondary cortex was 8 to 10 layered thick and made up of circular to isodiametric cells, the cells were thin-walled parenchyma. Many of these cells contained starch grains, orange brown contents and rosette crystals. The starch grains were mostly simple and shapes were circular to oval. This region also showed the presence of stone cells found in a singles or in groups, they were of two types i) thin walled and broad lumened with simple pits on their walls and ii) heavily thickened walled with prominent striations and narrow lumened with simple pits on their walls. The sclereids and mucilage canals were also found scattered in this region. Many sclereids showed the brown depositions. The inner bark consisted of phloem tissue, bast fibres and medullary rays. The phloem parenchyma cells were thin walled, many of them contained orange brown contents, few spherical starch grains and rosette crystals. Alternating with the parenohymatous elements were also found the presence of fibres, mostly in groups. The fibers were thick walled, narrow lumened and pointed tips. The medullary rays were heterogeneous (3 to 6 seriate). The cells were radially elongated and fairly thick-walled, most of them filled with rosette crystals while few with starch grains.

Bark : Powder study (Fig. 142.)

The components present in the powder were thin walled cork cells, parenchyma containing orange brown contents, rosette crystals, spherical starch grains, thin walled ,broad lumened stone cells, narrow lumened heavily thickened stone cells with striations, sclereids with brown depositions, phloem ray containing rosette crystals, thick walled narrow lumened fibers.

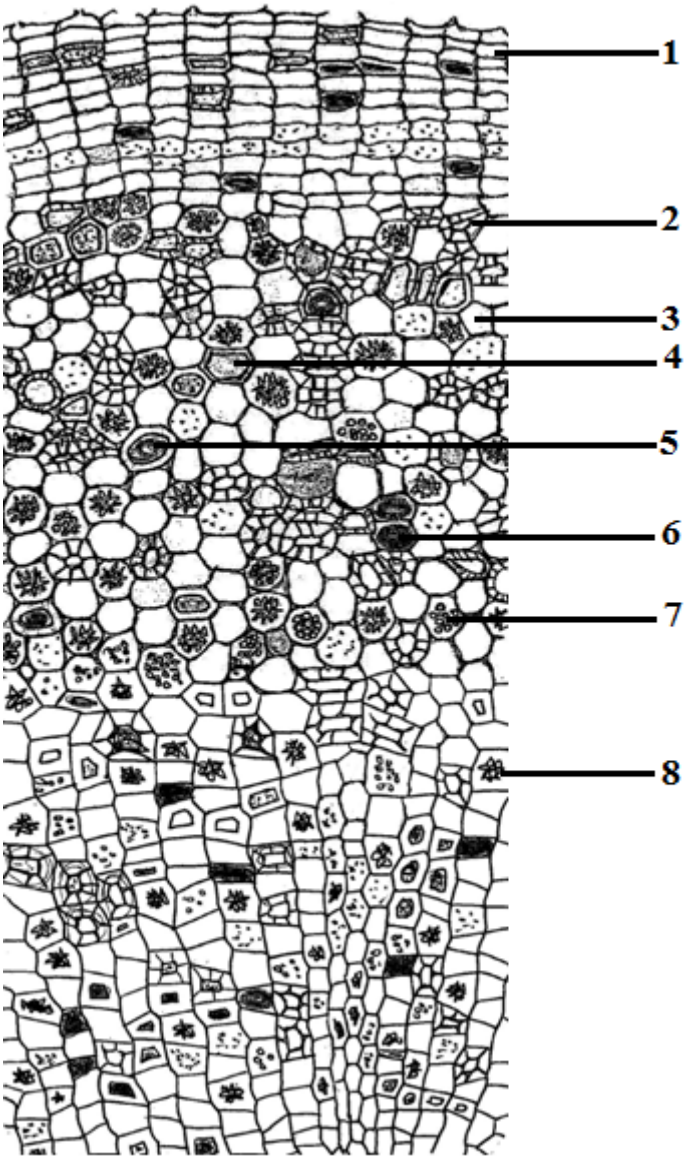


Fig. 141. ***Bombax ceiba*** **bark, T.S**: 1.Thin walled cork cells, 2. Stone cell, 3.Secondary cortex, 4. Sclereids, 5. Mucilage cells, 6. Parenchyma containing orange brown contents, 7.Starch grains, 8.Rosette crystals.

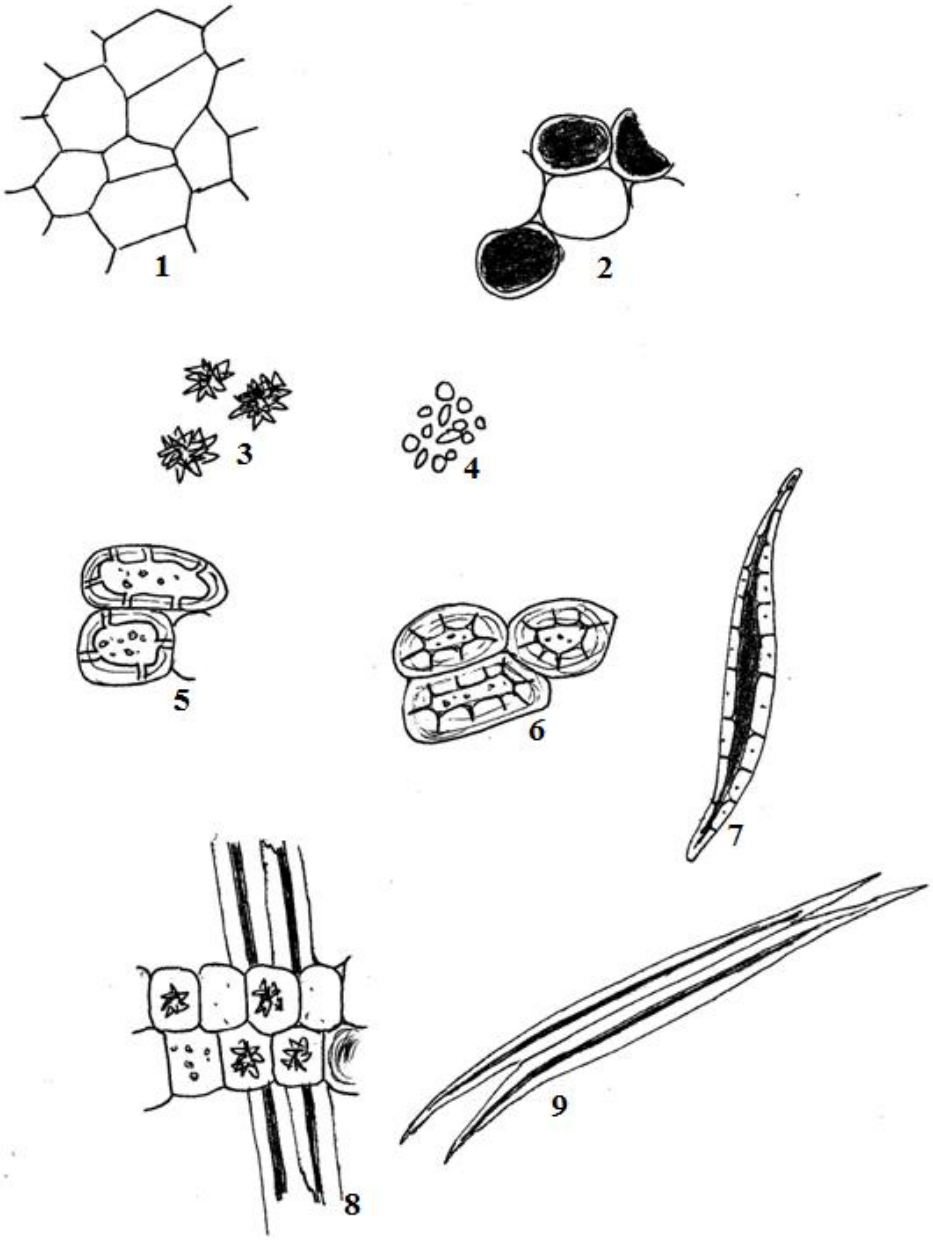


Fig. 142. ***Bombax ceiba*** **bark, powder study**: 1.Thin walled cork cells, 2.Parenchyma containing orange brown contents, 3. Rosette crystals, 4.Spherical starch grains, 5.Thin walled ,broad lumened stone cells, 6. Narrow lumened heavily thickened stone cells, 7.Sclereids with brown depositions, 8. Phloem ray containing rosette crystals, 9.Thick walled narrow lumened fibres.

Distinguishing features

Phytochemical markers

1. Cyanidin.
2. Vanillic acid.
3. Syringic acid.
4. Ferulic *(cis*- and *trans*- isomers) acid.
5. Rhamnose.
6. Galactose.
7. Arabinose.
8. Glucose.

Pharmacognostic markers

1. Thin walled cork cells.
2. Parenchyma containing orange brown contents.
3. Starch grains.
4. Stone cells.
5. Sclereids with brown depositions.
6. Rosette crystals.
7. Absence of crystal fibres.
8. Rosette crystals present in the medullary rays.

Physico-chemical analysis:

Table : Values obtained for the proximate analysis.

Sr.No.	Parameter	Mean ± SD (%)*			Average (%)
		Summer	Monsoon	Winter	
1.	Total Ash Content	8.74±0.53	8.69±0.27	8.99±0.31	8.81
2.	Acid Insoluble Ash content	1.23±0.11	1.09±0.19	1.66±0.03	1.33
3.	Alcohol soluble extractive	7.44±0.28	6.83±0.27	6.86±0.31	7.04
4.	Water soluble extractive	11.43±0.19	11.01±0.04	10.93±0.17	11.12

*Each value is a mean of 3 reading.

7.d.*Polyalthia longifolia* Benth (Sonn.) Thwaites (Annonaceae)

Sanskrit: Ashwapallava, Kasthadaru.

Vernacular names

Bengali: Debdaru.

English : Cemetery Tree.

Gujarati : Asopalav.

Hindi: Asoka, Debdari.

Kannada: Kambadarnara, Hessare.

Malayalam: Arena, Chorana.

Oriya: Asupal, Debdaru.

Tamil : Nettaingam, Assothi.

Telugu: Nara maamidl.

Distribution and habitat

The plant is an evergreen shrub or tree grows throughout the tropical and subtropical parts of India up to an altitude of 1500 m.

Morphological features

The plant showed the slender branches, short, about 1-2 m long, glabrous, and pendulous. Leaves alternate, exstipulate, distichous, mildly aromatic, 7.5-23 by 1.5-3.8 cm, shining, glabrous, narrowly lanceolate, tapering to a fine acuminate apex, margin markedly undulate, pinnately veined, leathery or subcoriaceous, shortly petiolate; petiole about 6 mm long. Flowers arise from branches below the leaves, nonfragrant, 2.5-3.5 cm across, yellowish to green, in fascicles or shortly pendunculate umbels; petals 6, 2 seriate, flat, from a broad base, lanceolate, long accumulate, spreading; and sepals 3, broad, short, triangular, the tips reflexed. Stamens many, cuneate; connective truncately dilated beyond the cells. Ovaries indefinite; ovules 1-2; style oblong. Ripe fruits ovoid, 1.8-2 cm long, numerous, stalked, glabrous, 1 seeded; stalk 1.3 cm long, short, glabrous. Seeds smooth, shining.

Medicinal uses

This plant has been used in traditional system of medicine for the treatment of fever,skin diseases, diabetes, hypertension and helminthiasis (Kirtikar and Basu 1995). The bark and leaves of this plant display effective antimicrobial activity, cytotoxic function and hypotensive effects (Katka, Suthar and Chauhan 2010).

Previous Phytochemical reports

The plant mainly contains diterpenoids, alkaloids, tannins, and mucilage. The chief components of the plant are O-methylbulbocapnine-*N*-oxide, polyfothine , *N*-methylnandigerine-*N*-oxide, oliveroline-*N*-oxide, pendulamine A, *N*-pendulamine B , 8-oxopolyalthiane, 16-oxo-5, 13-halimadien-15-oic acid , 16-Oxo-3, 13-clerodadien-15-oic acid, 16-hydroxycleroda-3, 13-dien-16, 15-olide.Two clerodane-type diterpenoids have been isolated and identified as 16α-hydroxy-cleroda-3,13(14)Z-dien-15,16-olide and 16-oxo-cleroda-3,13 (14)E-dien-15-oic acid on the basis of spectral properties. A γ-methoxybutenolide clerodane diterpene 2 has been isolated from the petroleum ether extract of the bark. Its structure has been deduced by spectral analyses and by chemical correlation with the corresponding γ-hydroxybutenolide diterpene 1, isolated earlier from this plant. Aporphine and azafluorene alkaloids, proanthocyanidins, h-sitosterol, and leukocyanidin, clerodane, and ent-helimane, diterpenoids were isolated from the leaves, stem, and stem bark. Carbohydrate was isolated from the seeds. A novel azafluorene alkaloid, polylongine (5-hydroxy-6-methoxy-1-methyl-4-azafluoren-9-ol), and 3 new aporphine *N*-oxide alkaloids named (+)-O-methylbulbocapnine-β-*N*-oxide, (+)-O-methylbulbocapnine-α-*N*-oxide, and (+)-*N*-methylnandigerine-β-*N*-oxide were isolated from the leaves. The leaf oil was almost exclusively composed of sesquiterpene derivatives, being represented by allo-aromadendrene (19.7%), caryophyllene oxide (14.4%), β-caryophyllene (13.0%), β-selinene (7.9%), α-humulene (7.0%) and ar-curcumene (6.8%). However, α-copaene and α-muurolol (approx 8.7%), β-selinene (8.6%), viridiflorene (8.1%), α-guaiene (7.8%), allo-aromadendrene (7.4%), and δ-cadinene (7.0%) were the major constituents in the oil of the bark.(Katka, Suthar and Chauhan 2010).

Previous pharmacognostic reports

Little data is available on pharmacognosy of the stem bark of this plant (Anon.1999 and Malati G and Pillai 2005).

Materials and methods

The plant material has been collected from Vadodara, Gujarat. Phytochemical analysis of bark of the plant for their secondary metabolites such as alkaloids, flavonoids, phenolics, steroids, quinones, mucilages and other compounds were done by standard methods of chromatography and spectrophotometry described in Chapter 2. Pharmacognostic, HPTLC finger printing and physico-chemical studies were done by using standard methods described in Chapter 2.

Results

Phytochemistry

The bark was found to contain proanthocyanidins and the phenolic acids were vanillic, syringic and ferulic (*cis*- and *trans*- isomers) acids. Mucilage amounted to 4.9 % consisting of rhamnose, xylose and glucose. The plant also showed the presence of alkaloids and steroids.

Pharmacognosy

Macroscopic characters (Fig.143.)

Stem bark was flat to curved, externally rough, orange-brown and showed faint ridges and furrows with vertical lenticels. The fracture was hard and fibrous.

Fig.143. *Polyalthia longifolia* bark.

Microscopic characters

Bark : T.S (Fig. 144)

The T.S. of stem bark showed the periderm composed of 5 to 12 rows of tangentially elongated cork cells. The cells were with light brown thick walls. The outer few rows of cork cells were much compressed. Some of these cells contain yellowish- brown contents. Isolated stone cells were seen in this region. The Phellogen was single layered. The cells of phelloderm were thin walled and tangentially elongated. There were few groups of rectangular stone cells were embedded in this region .The simple rounded starch grains and prismatic crystals were also found present along with few cells containing light brown masses. The secondary cortex was very narrow composed of thin walled parenchymatous cells. The cells were polygonal to isodiametric shaped and compactly arranged with several prominent groups of sclereids, stone cells, mucilage canal and oilc ells embedded in this region. Starch grains, prismatic crystals and brown contents were also occurred in many of the cortical parenchyma. The presence of plenty of acicular crystals was characteristic. Very few stone cells found here were thickened walled, narrow lumened with prominent striations and simple pits on their walls. The phloem consisted of soft tissue, bast fibres and medullary rays. The phloem parenchyma cells were small thin walled, many of them contained brown contents , rounded starch grains, acicular and prismatic crystals. There were groups of fibres distributed in this region forming a discontinuous ring. The fibers were thickened walled narrow lumened and both sepetate and asepetate types. The stone cells present in this region were comparatively smaller than that of cortex and were in traces. The medullary rays were multi-seriate. The cells of the medullary rays were mostly filled with acicular and prismatic crystals while starch grains were found in intraces.

Bark : Powder study (Fig. 145)

The components present in the powder were thick walled cork cells, parenchyma with brown contents and starch grains, prismatic and acicular crystals, parenchyma with acicular crystals, sepetate and asepetate fibres.

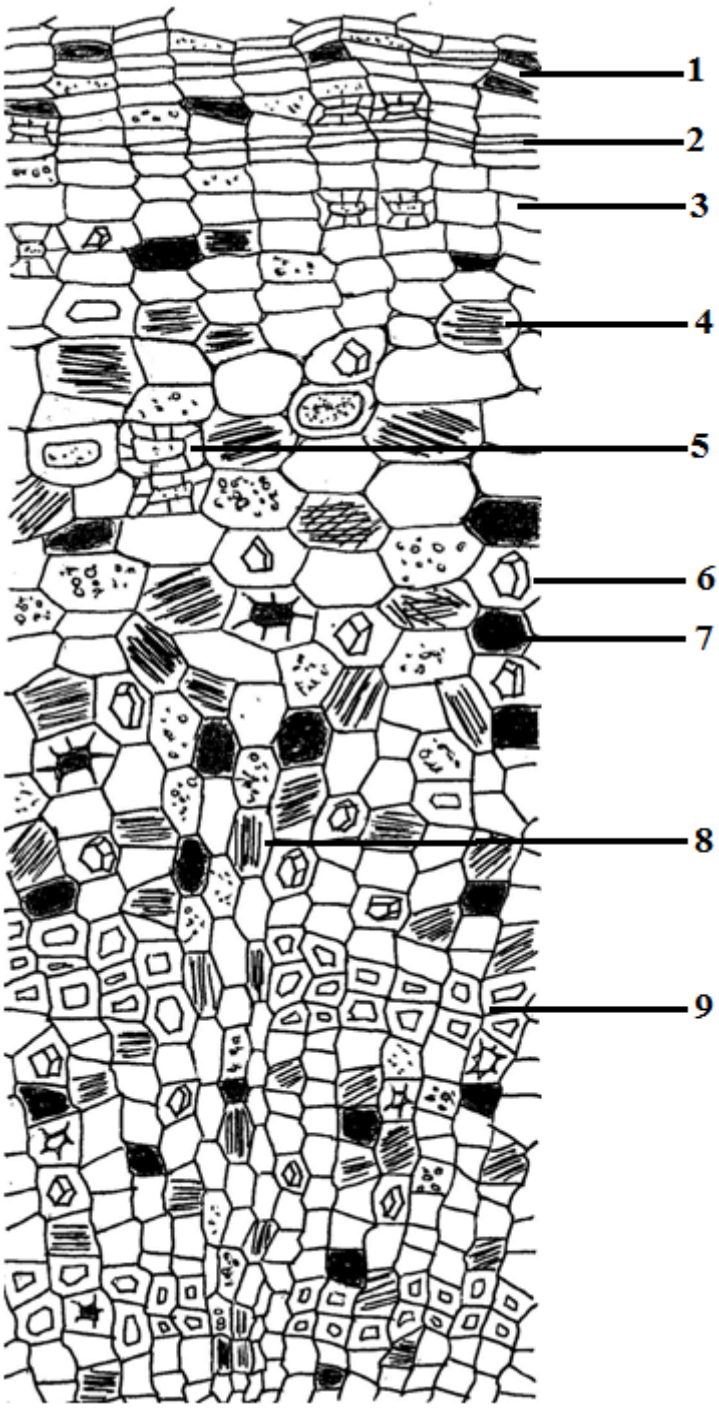


Fig. 144. ***Polyalthia longifolia*** **bark, T.S:** 1.Cork, 2. Phellogen, 3. phelloderm, 4.Parenchyma with acicular crystals, 5. Stone cells, 6. Prismatic crystals, 7. Parenchyma with brown deposits, 8. Phloem rays, 9. Phloem fibers.

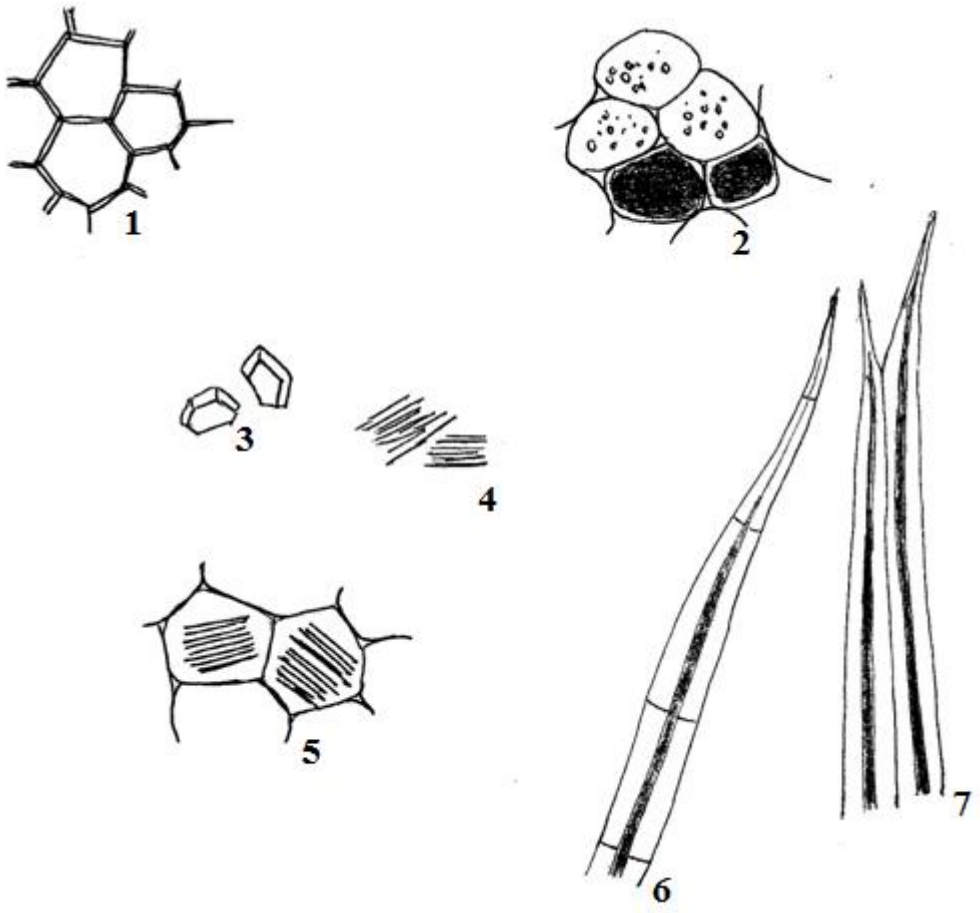


Fig. 145. ***Polyalthia longifolia*** **bark, powder study:** 1.Cork, 2. Parenchyma with brown contents and starch grains,3. Prismatic crystals, 4. Acicular crystals, 5. Parenchyma with prismatic crystals, 6. Sepetate fibers, 7. Asepetate fibers.

Distinguishing features

Phytochemical markers

1. Proanthocyanidins.
2. Vanillic acid.
3. Syringic acid.
4. Ferulic (*cis*- and *trans*- isomers) acid.
5. Rhamnose.
6. Xylose.
7. Glucose.

Pharmacognostic markers

1. Thick walled cork cells.
2. Parenchyma containing brown contents.
3. Starch grains.
4. Acicular crystals.
5. Septate and aseptate fibres.
6. Absence of crystal fibres.

Physico-chemical analysis:

Table: Values obtained for the proximate analysis.

Sr.No.	Parameter	Mean ± SD (%)*			Average (%)
		Summer	Monsoon	Winter	
1.	Total Ash Content	7.29±0.42	7.66±0.39	7.56±0.16	7.50
2.	Acid Insoluble Ash content	0.79±0.48	0.86±0.27	0.87±0.03	0.84
3.	Alcohol soluble extractive	17.37±0.41	16.37±0.44	16.56±0.28	16.77
4.	Water soluble extractive	19.09±0.33	19.29±0.19	19.33±0.51	19.24

*Each value is a mean of 3 readings.

7.e.*Shorea robusta* Gaertn. (Dipterocarpaceae)

Sanskrit: Agnivallabha, Asoka, Asvakarna, Vansha, Salah Ashvakarna, , Sarja.

Vernacular names:

Assamese : Sal.

Bengali*:* Shaalgaach.

English*:* Saltree, Shaal tree.

Gujrati*:* Shaal.

Hindi : Saal, Sakhuaa, Saakhu.

Kannada : Kabba, Saal

Malayalam:Saalvriksham,

Mulappumarutu

Marathi:Shaalvriksh

Oriya : Salva, Shaaluaagachha.

Punjabi : Shala.

Tamil : Saalam.

Telugu : Guggilam.

Distribution and habitat

The plant is large sub-deciduous tree, found extensively in parts of North-East and Central India.

Morphological features

The plant is 18-30 m in height. The leaves simple, ovate-oblong, acuminate, tough, coriaceous, glabrous, base cordate or rounded, lateral nerves 12-15 pairs; flowers yellowish, in axillary or terminal panicles, stamens upto 50, connectives with subulate bearded appendages, minutely 3-fid at the apex; fruits indehiscent, ovoid with 5 equal wings; seeds ovoid with fleshy unequal cotyledons.

Medicinal uses:

The bark and leaves are astringent, acrid, cooling, anthelmintic, alexeteric, anodyne, constipating, urinary astringent, union promoter, depurative and tonic. They are useful in vitiated conditions of kapha and pitta, ulcers, wounds, bacterial affections, diarrhoea, dysentery, gonorrhea, leucorrhoea, pruritus, leprosy, cough, hyperhidrosis , haemorrhoids and anaemia. The fruits are astringent, cooling, aphrodisiac, cholagogue and tonic, and are useful in dipsia, burning sensation,

tubercular ulcers, seminal weakness and dermatopathy. The resin is cooling, anodyne, vulnerary, antibacterial, deodorant, constipating, detergent, carminative, stomachic, aphrodisiac,expectorant, ophthalmic and tonic. It is useful in hypearhidrosis, vitiated conditions of pitta, wounds, ulcers, neuralgia, burns, pruritus, fever,diarrhea, dysentery, haemorrhoids, gonorrhea, menorrhagia, splenomegaly, obesity, cephalalgia, odontalgia, burning of the eyes and ophthalmodynia.(Warrier *et.al*.,1994).

Previous Phytochemical reports

The plant shows the presence of ursolic acid and *α*-amyrenone; α & β-amyrin **(** Hota and Bapuji, 1993, Mishra and Ahmed. 1997**);** bark contains ursonic acid and oleanane, Shoreaphenol (Harbone1999; Patra *et.al*.,1992)**;** seed contains hopeaphenol, leucoanthocyanidin, and 3,7-dihydroxy-8-methoxyflavone7-*O*-*α*-l-rhamnopyranosyl-(1→4)-*α*-l- rhamnonopyrano- syl(1→6)-*β*-d-glucopyranoside (Prakash and Rao. 1999)**;** while heartwood contains germacrene-D (Kaur *et.al* .,2001). The isolation of *β*-amyrin, friedelin, *β*-sitosterol, pheophytin-*α*, and dihydroxyisoflavone from mature leaves was also reported (Chauhan *et.al*., 2002).

Previous pharmacognostic reports

No study has been done on the pharmacognostic characters of the bark of this plant.

Materials and methods

The plant material has been collected from Madhya Pradesh. Phytochemical analysis of bark of the plant for their secondary metabolites such as alkaloids, flavonoids, phenolics, steroids, quinones, mucilages and other compounds were done by standard methods of chromatography and spectrophotometry described in Chapter 2. Pharmacognostic, HPTLC finger printing and physico-chemical studies were done by using standard methods described in Chapter 2.

Results

Phytochemistry

The stem bark is found to contain flavonid, 3- OMe quercetin and anthocyanin pelargonidin derivatives. The phenolic acids were vanillic, syringic and *p*- hydroxy benzoic acids. Mucilage amounted to 5.2% consisting of rhamnose and glucose. The plant also showed the presence of steroids.

Pharmacognosy

Macroscopic characters (Fig.146)

Stem bark was curved , externally rough, yellowish brown and with deep prominent longitudinal fissures .The fracture was hard and fibrous.

Fig.146.*Shorea robusta* bark.

Microscopic characters

Bark : T.S (Fig. 147)

The T.S. of stem bark showed the periderm composed of 8 to 18 rows of squarish to slightly tangentially elongated cork cells. The cells showed the light brown thick walls. The outer few rows of cork cells were much compressed and their cell walls were wavy. Some of these cells contained reddish brown contents. The Phellogen was indistinct. The phelloderm made up of polygonal to isodiametric shaped cells and the walls were thin, interspersed in which were rhomboidal crystals , starch grains and sclereids. The middle bark which was mainly secondary cortex was composed of thin walled parenchymatous cells. The cells were polygonal to isodiametric shaped and compactly arranged with several prominent groups of sclereids. A few small spherical starch grains were also occurred in most of the parenchyma cells, while others contain rhomboidal crystals of calcium oxalate of various sizes and reddish brown contents. Abundant stone cells with various shapes were found present in the phelloderm and secondary cortex region and were with thick walled, broad lumened with distinct striations and pit canals. These region also showed the presence of few gum ducts as long tangential bands. The phloem consisted of sieve tubes, bast fibres and medullary rays. The phloem parenchyma cells were small thin walled, many of them contained reddish brown contents,

spherical starch grains and rhomboidal crystals. Alternating with the parenchymatous elements were also found the presence of fibres, mostly in groups of two. The fibers were aseptate type and found associated with rhomboidal crystals. The medullary rays were heterogeneous (3 to 7 seriate) and most of them filled with starch grains. The cells were radially elongate at outer and appeared tangentially elongated in the inner.

Bark : Powder study (Fig. 148)

The components present in the powder were thick walled cork cells with wavy walls, spherical starch grains, thick walled stone cells with broad lumen, sclereids, gum ducts, rhomboidal crystals, crystal fibres.

Distinguishing features

Phytochemical markers

1. 3- OMe Quercetin.
2. Pelargonidin derivatives.
3. *p*- Hydroxy benzoic acid.
4. Vanillic acid.
5. Syringic acid.
6. Glucose.
7. Xylose.

Pharmacognostic markers

8. Thick and wavy walled cork cells.
9. Parenchyma containing spherical starch grains.
10. Thick walled stone cells with broad lumen, distinct striations and pit canals .
11. Sclereids.
12. Gum ducts.
13. Rhomboidal crystals.
14. Heterogeneous medullary rays filled with starch grains
15. Crystal fibres.

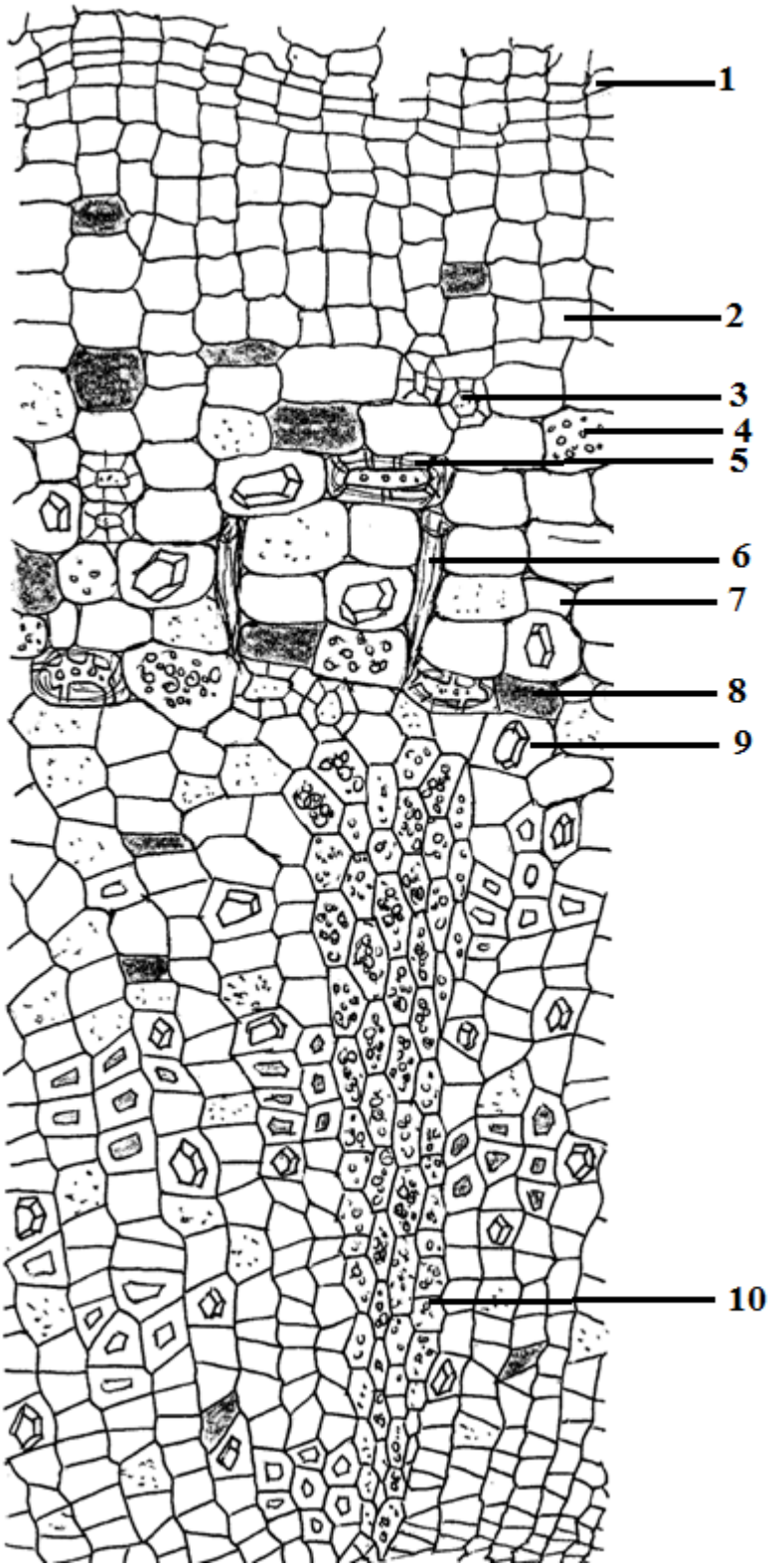


Fig.147. *Shorea robusta* bark, T.S:1.Cork, 2.Phelloderm, 3.Sclereids, 4.Parenchyma with starch grains, 5. Stone cells, 6. Gum duct, 7. Secondary cortex, 8. Parenchyma with reddish brown deposites, 9. Rhomboidal crystal, 10.Phloem rayes.

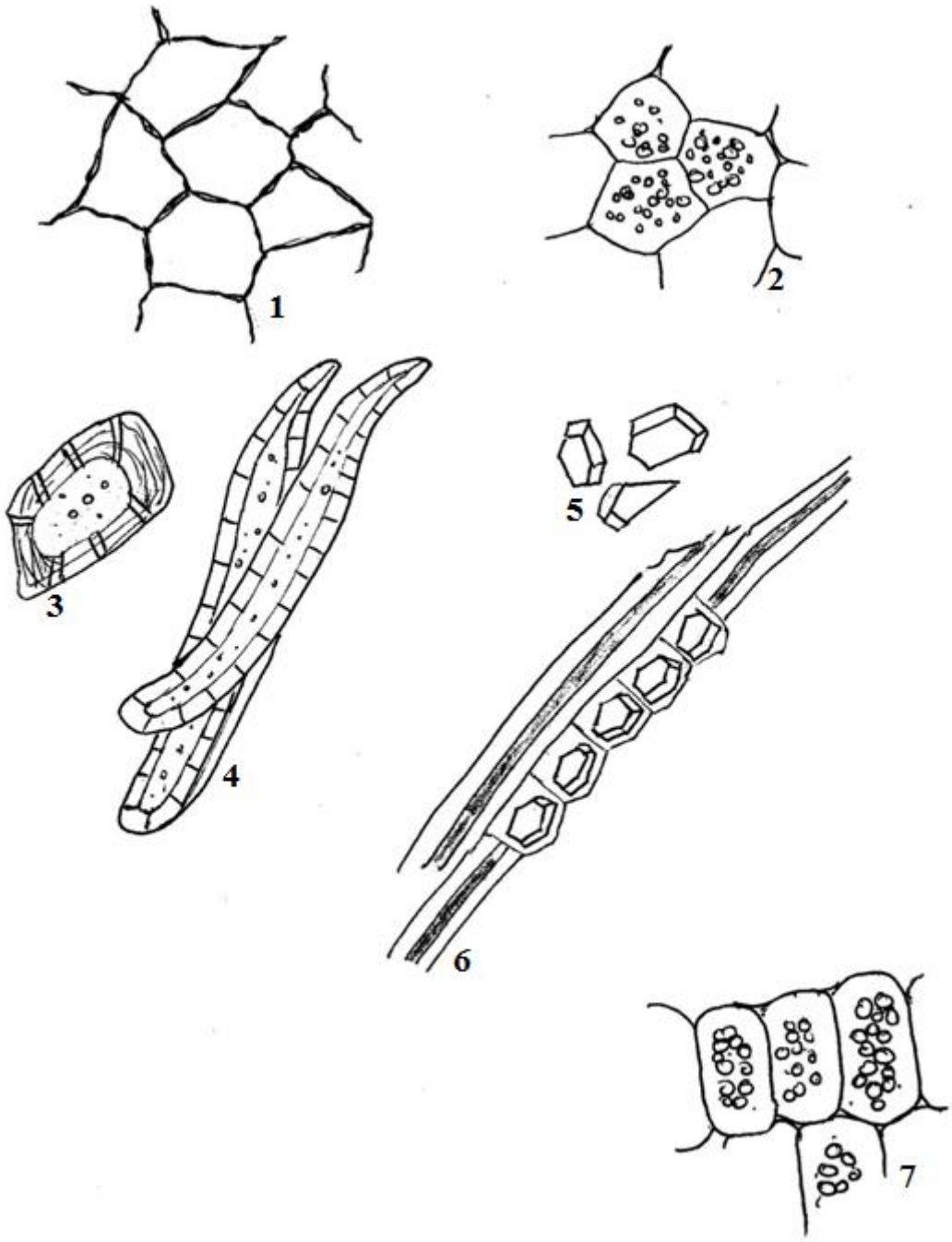


Fig. 148.*Shorea robusta* bark, powder study: 1.Thick walled cork cells, 2.Parenchyma containing spherical starch grains, 3.Stone cells, 4.Sclerides, 5.Rhomboidal crystals, 6.Crystal fibres, 7. Phloem ray with starch grains.

Physico-chemical analysis:

Table : Values obtained for the proximate analysis.

Sr.No.	Parameter	Mean ± SD (%)*			Average (%)
		Summer	Monsoon	Winter	
1.	Total Ash Content	9.12±0.37	9.19±0.13	9.29±0.16	9.2
2.	Acid Insoluble Ash content	0.99±0.29	0.99±0.33	1.03±0.17	1.00
3.	Alcohol soluble extractive	11.59±0.54	11.20±0.49	11.39±0.29	11.39
4.	Water soluble extractive	12.18±0.33	12.01±0.21	12.09±0.34	12.09

*Each value is a mean of 3 readings.

7.f. *Trema orientalis* (L.) Blume (Ulmaceae)

Sanskrit: Jivanti, Swahili.

Vernacular names:

Bengali : Jiban

English : Charcoal tree, Pigeon wood, Indian nettle tree, Indian charcoal tree.

Gujarati : Gol.

Hindi : Jivan, Jivanti Kshayanashini, Parvati Pranak

Konkani : Khargul

Malayalam : Amaraaththi, Aamaththaali, Pottaama, Pottaamaram

Marathi : Gol, Kapshi Khargol

Sanskrit : Jivani, Jivanti Pranaka

Tamil : Pey-Munnai

Telugu : Boggu Chettu, Charapappuchettu, Morali Pruyalavriqshamu.

Distribution and habitat

This is an evergreen shrub or tree up to 18 m in height found in the lowland humid tropics. It has a very wide distribution, occurring from tropical Africa southwards to South Africa , and eastwards to southern Asia.

Morphological features

The plant is found with heavy branching and rounded to spreading crown. The slender branchlets are covered with white velvety hairs. Leaves simple, alternate, stipulate, along drooping branches, to 14 cmlong, papery, rough to the touch and dull above, short grey hairs below,the edge finely toothed all round, blade unequal sided.Flowers small, green or greenish-white, unisexual, borne in a crowdedinflorescence consisting mainly of male flowers with a few female ones atthe top.Fruit small, round and fleshy,glossy black when ripe, 4-6 mm, containing1 dull black seed embedded in bright green flesh.

Medicinal uses

The root of the plant is used in the treatment of diarrhoea, asthma and passing of blood in urine; the bark is used as poultic in muscular pain; the roots, barks and leaves are used in epilepsy. In African folk medicine, it is used in many diseases including cough, dysentery, hypertension, asthma.(Chowdhury and Islam,2004).It is used in the treatment of diabetes mellitus, respiratory diseases, oliguria, and malaria (Adinortey *et.al*.,2013).

Previous Phytochemical reports

The bark showed the presence of methylswertianin, decussatin, glycosides of decussatin, sweroside, scopoletin, (-)-epicatechin, lupeol, p -hydroxybenzoic acid, 3,4-dihydroxybenzoic acid, adian-5-en-3-one, 2a, 3a, 23-trihydroxyurs-12-en-28-oic acid, 2a, 3ß-dihydroxyurs-12-en-28-oic acid, ß-sitosterol, 3- O -ß-glucopyranosyl-ß-sitosterol and hexacosanoic acid.

(Tchamo *et.al.*,2001).

Previous pharmacognostic reports

No study has been done on the pharmacognostic characters of the stem bark of this plant.

Materials and methods

The plant material has been collected from Pavagadh, Gujarat. Phytochemical analysis of bark of the plant for their secondary metabolites such as alkaloids, flavonoids, phenolics, steroids, quinones, mucilages and other compounds were done by standard methods of chromatography and spectrophotometry described in Chapter 2. Pharmacognostic, HPTLC finger printing and physico-chemical studies were done by using standard methods described in Chapter 2.

Results

Phytochemistry

Along with reported scopoletin the bark was found to contain high amount of anthocynidin cyanidin. The phenolic acids were vanillic, syringic and *p*- hydroxy benzoic acids. Mucilage amounted to 5.2% consisting of rhamnose and glucose. The plant also showed the presence of alkaloids, iridoids and steroids.

Pharmacognosy

Macroscopic characters (Fig.149)

Stem bark was channeled , externally smooth, orange-brown and showed longitudinal wavy wrinkles and transverse corky spots. The fracture was short and fibrous.

Fig. 149. *Trema orientalis* bark.

Microscopic characters

Bark : T.S (Fig. 150)

The T.S. of stem bark showed the periderm composed of 5 to 10 rows of squarish to tangentially elongated cork cells. The cells showed the light brown thick walls. The outer few rows of cork cells were much compressed. Some of these cells contain yellowish- brown contents. There were few rectangular stone cells found present in this region. The Phellogen was single or two layered. The cells of phelloderm were almost squarish and little larger than cork cells and the walls were thin, interspersed in which were rhomboidal crystals , starch grains and sclereids. The secondary cortex was composed of thin walled parenchymatous cells. The cells were polygonal to isodiametric shaped and compactly arranged with several prominent groups of sclereids and stone cells. Rosette crystals of calcium oxalate and spherical starch grains were also occurred in many of the cortical parenchyma. The presence of yellow or yellowish brown contents in these cells were very common. Characteristically the stone cells were radially arranged one above the other, in various shapes from cubical to linear and of two types i) thin walled and broad

lumened with simple pits on their walls and ii) heavily thickened walled with prominent striations and narrow branched lumened with simple pits on their walls. The presence of yellow or yellowish brown contents in stone cells and sclereids were characteristics. The phloem consisted of phloem tissue, bast fibres and medullary rays. The phloem parenchyma cells were small thin walled, many of them contained yellowish brown contents, spherical starch grains and rosette crystals. There were groups of fibres distributed in this region. The fibers were of two types i) thin walled and broad lumened and ii) thickened walled narrow lumened. The medullary rays were mostly bi-seriate. Few of them filled with starch grains and with yellowish brown contents.

Bark : Powder study (Fig. 151)

The components present in the powder were thick walled cork cells, parenchyma with starch grains, stone cells and sclerides with yellowish brown contents, parenchyma containing yellow contents and rosette crystals, thin and thick walled fibres.

Distinguishing features

Phytochemical markers

1. Scopoletin.
2. Cyanidin.
3. Vanillic acid.
4. Syringic acid.
5. *p*- Hydroxy benzoic acid.
6. Iridoids.
7. Rhamnose.
8. Glucose.

Pharmacognostic markers

1. Thick walled cork cells.
2. Parenchyma containing yellow contents.
3. Thin and thick walled stone cells laid as rack.
4. Deposition of yellowish brown contents in stone cells and sclereids.
5. Rosette crystals.
6. Thin and thick walled fibres.
7. Absence of crystal fibres.

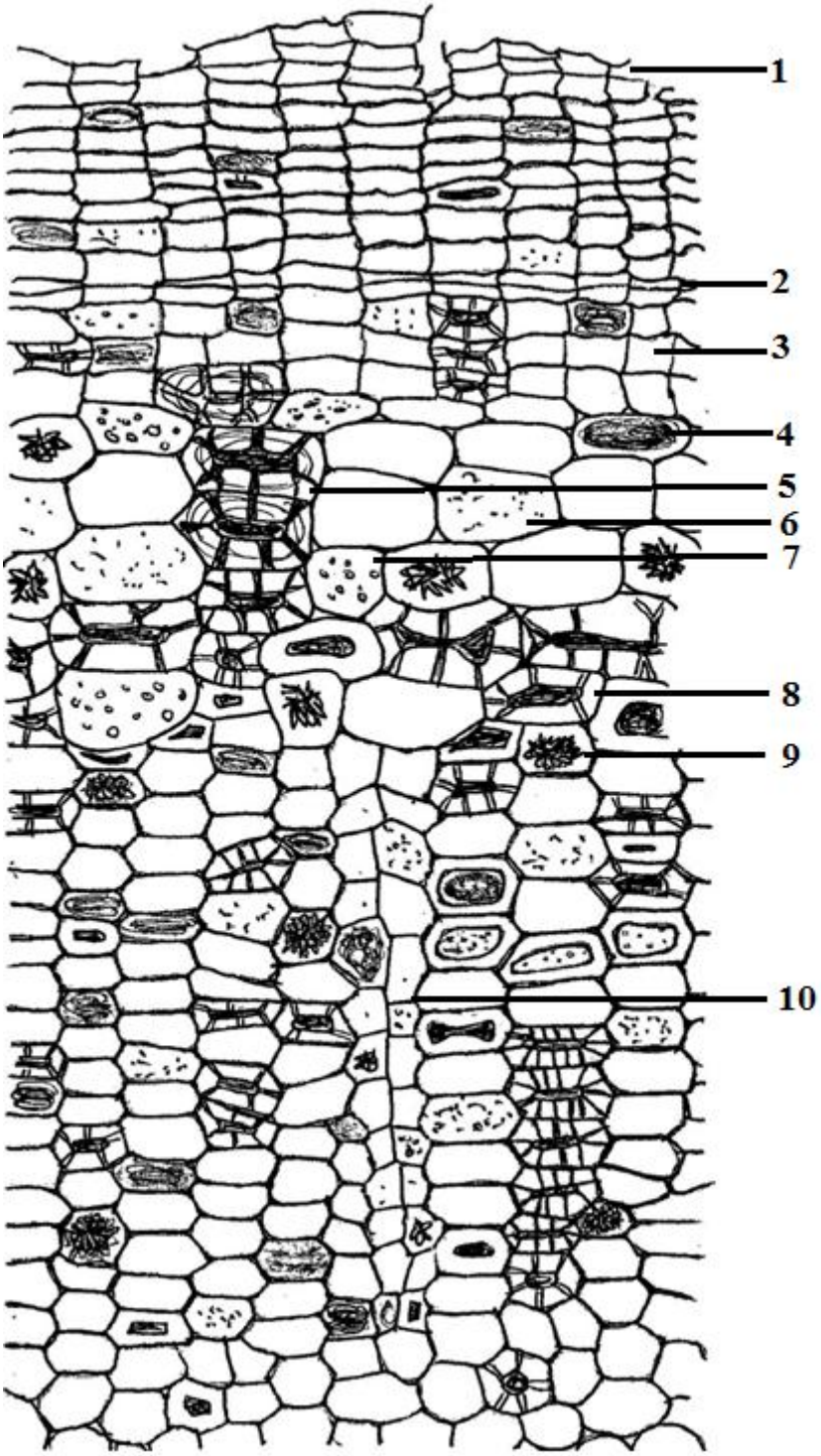


Fig. 150. *Trema orientalis* bark, T.S: 1.Cork, 2. Phellogen ,3. Phelloderm, 4. Parenchyma with yellowish brown deposits, 5. Stone cells, 6. Secondary cortex, 7. Starch grains, 8. Sclerides, 9. Rosette crystal, 10.Phloem rays.

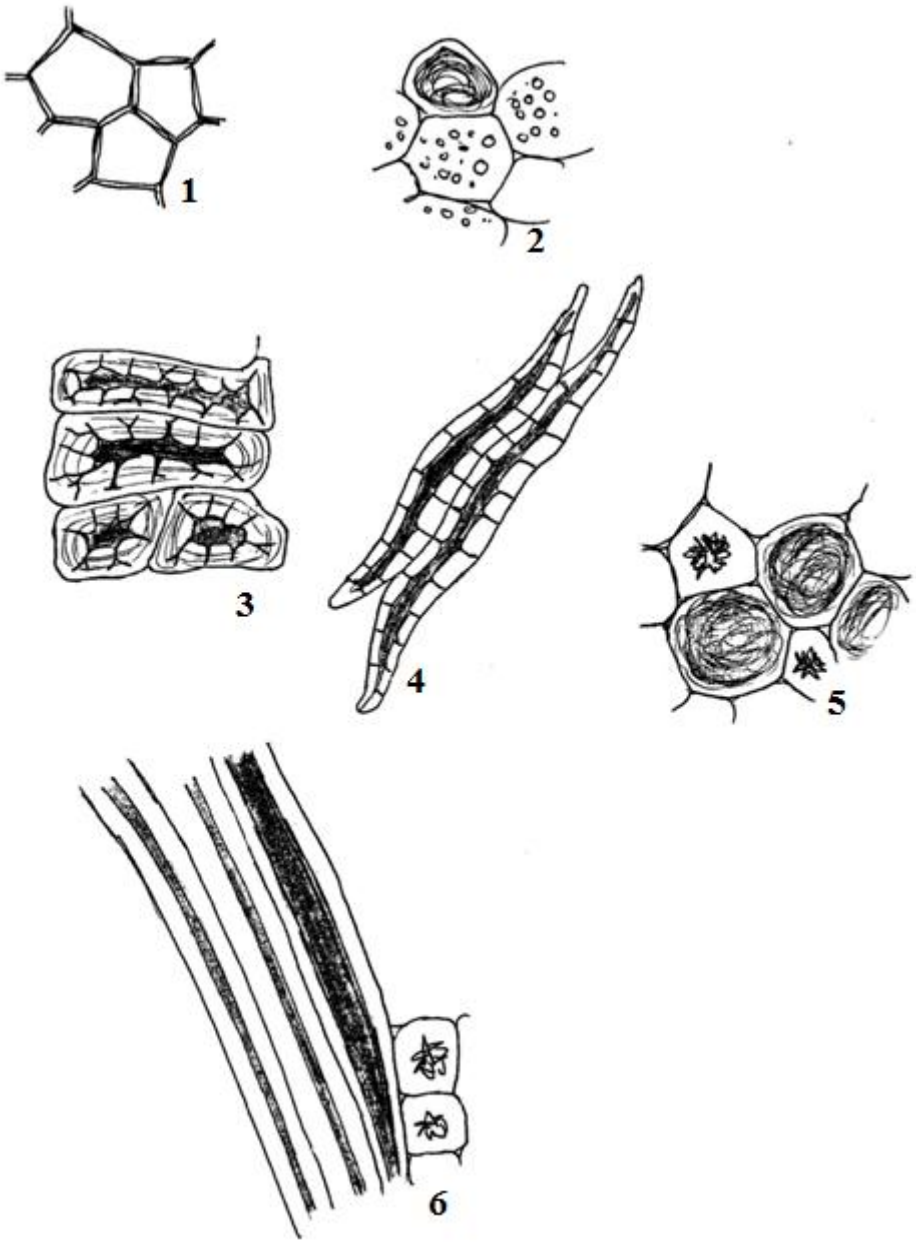


Fig. 151. *Trema orientalis* bark, powder study: 1.Cork, 2. Parenchyma with starch grains and yellowish brown deposits, 3. Stone cells with yellowish brown contents ,4. Sclereids, 5. Parenchyma with rosette crystals, 6. Thin and thick walled fibers.

Physico-chemical analysis:

Table : Values obtained for the proximate analysis.

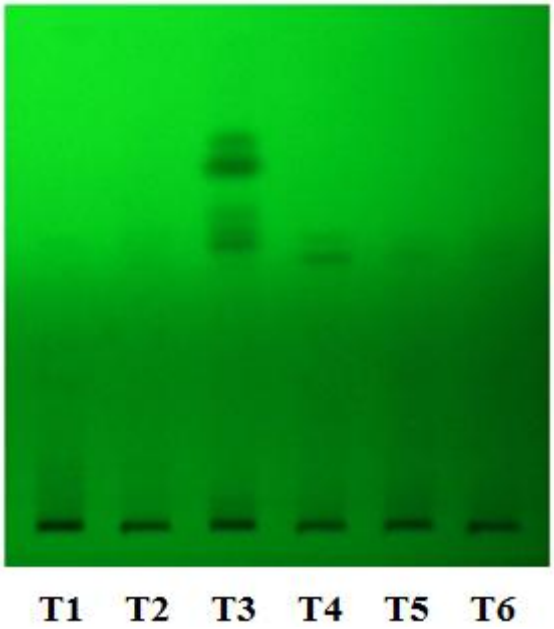
Sr.No.	Parameter	Mean ± SD (%)*			Average (%)
		Summer	Monsoon	Winter	
1.	Total Ash Content	4.13±0.29	4.08±0.31	4.14±0.29	4.12
2.	Acid Insoluble Ash content	0.89±0.21	0.86±0.19	0.87±0.22	0.87
3.	Alcohol soluble extractive	11.27±0.43	10.68±0.37	10.87±0.39	10.94
4.	Water soluble extractive	15.17±0.33	15.04±0.28	15.00±0.36	15.07

*Each value is a mean of 3 readings.

7.g. HPTLC fingerprinting and Physo-chemical analysis of *Fumaria parviflora* and its substitutes/adulterants

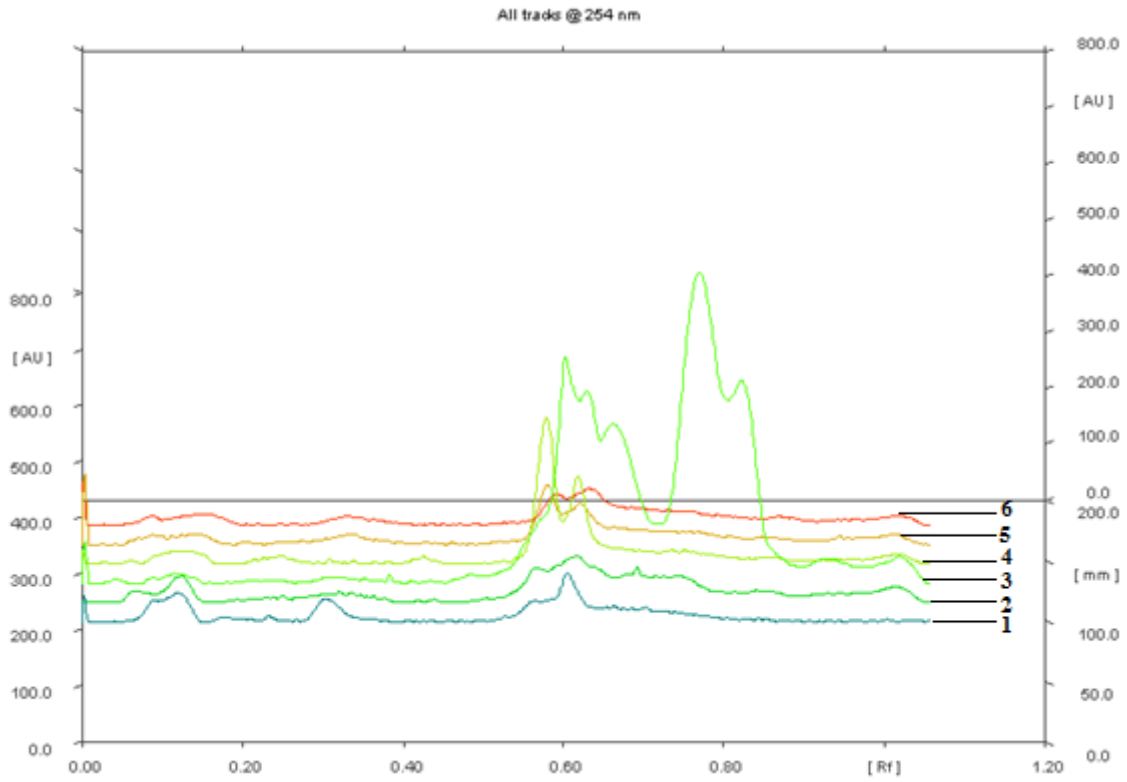
HPTLC fingerprinting

Figure 152.a: HPTLC chromatogram of *Saraca indica* and its substitutes/adulterants (UV 254 nm).



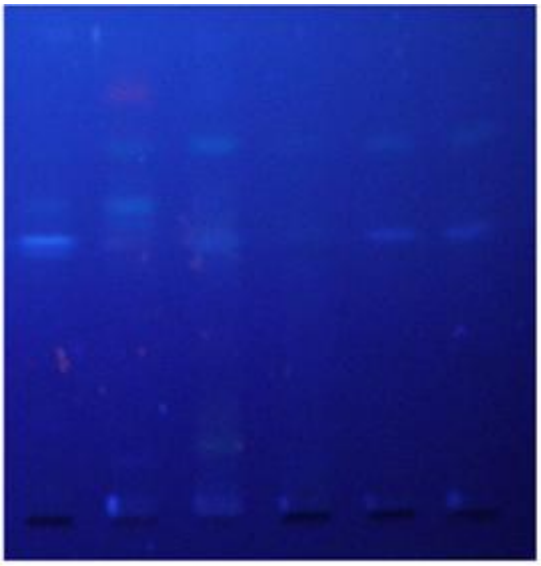
(a).T1-*Saraca indica*, T2-*Trema orientalis*, T3-*Polyalthia longifolia*, T4-*Bauhinia variegata*, T5-*Bombax ceiba*, T6-*Shorea robusta*.

Figure 152.b: HPTLC chromatogram of *Saraca indica* and its substitutes/adulterants (UV 254 nm).



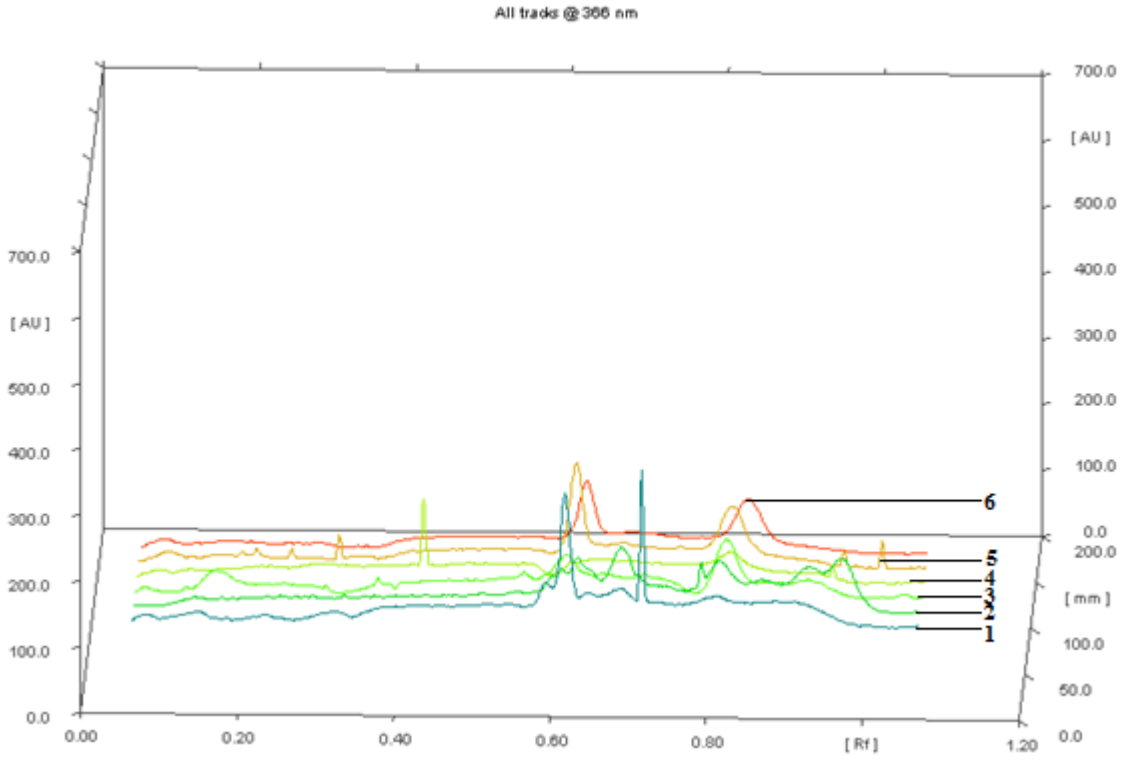
(b).1-*Saraca indica*, 2-*Trema orientalis*, 3-*Polyalthia longifolia*, 4-*Bauhinia variegata*, 5-*Bombax ceiba*, 6-*Shorea robusta*.

Figure 153.a : HPTLC chromatogram of *Saraca indica* and its substitutes/adulterants.(UV 366 nm).



(a).T1-*Saraca indica*, T2-*Trema orientalis*, T3-*Polyalthia longifolia*, T4-*Bauhinia variegata*, T5-*Bombax ceiba*, T6-*Shorea robusta*.

Figure 153.b : HPTLC chromatogram of *Saraca indica* and its substitutes/adulterants(UV 366 nm).



(b).1-*Saraca indica*, 2-*Trema orientalis*, 3-*Polyalthia longifolia*, 4-*Bauhinia variegata*, 5-*Bombax ceiba*, 6-*Shorea robusta*.

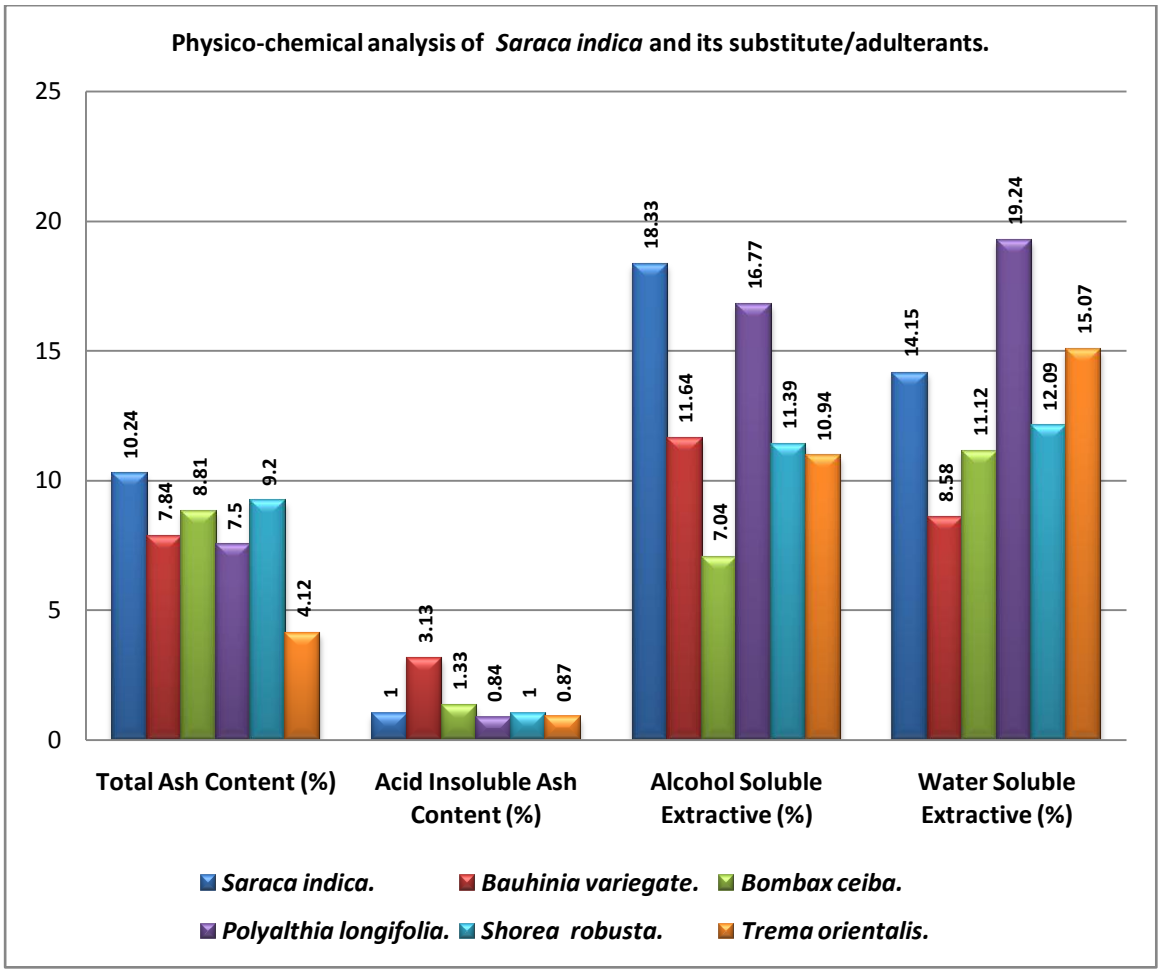
HPTLC profile of *Saraca indica* showed the presence of 5 peaks when observed under UV 254 nm (figure-152.a)and 9 peaks under 366 nm (figure-153.b). There were 4 major peaks found at R_f 0.12, R_f 0.30, R_f 0.56 and R_f 0.61 under UV 254 and 9 peaks at R_f 0.61, R_f 0.68 , R_f 0.70, R_f 0.80 and R_f 0.90 under 366 nm. That of *Trema orientalis* and *Bauhinia variegata* showed the presence of 7 peaks , *Bombax ceiba* and *Shorea robusta* 6 peaks and *Polyalthia longifolia* 11 peaks when observed under UV 254 nm.Under UV366 nm, *Trema orientalis*, *Polyalthia longifolia*, *Bauhinia variegata*, *Bombax ceiba* and *Shorea robusta* showed 8,7, 5, 9 and 3 peaks respectively.

HPTLC profile of *S. indica* and its substitutes/adulterants observed under UV 254 nm showed that *P. longifolia* was similar in 2 peaks but differed in 9 peaks. *T.orientalis*, *B. ceiba* and *S. robusta* were similar in 1 peak but *T. orientalis* differed in 6 peaks and *B. ceiba* and *Shorea robusta* in 5 peaks, while *B. variegata* did not show any peak similar to that of *S. indica* but differed in having 7 peaks.

HPTLC profile of *S. indica* and its substitutes/adulterants observed under UV 366 nm showed that *T. orientalis* was similar in 3 peaks but differed in 5 peaks. *B. ceiba* and *P. longifolia* were similar in 1 peak but differed *B. ceiba* in 8 peaks and *P. longifolia* in 6 peaks, while *B. variegata* and *S. robusta* did not show any peak similar to that of *S.indica* but *B. variegata* and *S. robusta* differed in having 5 and 3 peaks respectively.

Physico-chemical analysis

Physico-chemical analysis of *Saraca indica* and its substitutes/adulterants.



Total ash content

Total Ash Content of *Saraca indica* (10.24 %) along the material collected in different season does not show significant variation (Table-1) while the closest value to the substitute/adulterant in descending order is 9.2% (*Shorea robusta*), 8.81% (*Bombax ceiba*), 7.84% (*Bauhinia variegata*), 7.50% (*Polyalthia longifolia*) and 4.12% (*Trema orientalis*).

Acid insoluble ash content

Acid insoluble ash content of *Saraca indica* (1.00 %) along the material collected in different season does not show significant variation (Table-1). The adulterant *Shorea robusta* had the same value (1.00%) as of *Saraca indica*, while the closest value to the other substitute/adulterant in descending order is 0.87% (*Trema orientalis*), 0.84%(*Polyalthia longifolia*), 1.33% (*Bombax ceiba*)and 3.13 % (*Bauhinia variegate*).

Amongst the substitutes/adulterants of *S. indica* , the *S. robusta* showed the closest value of total ash content which showed that the *S. robusta* was more close to *S. indica* as compared to other substitutes/adulterants of *S. indica* .

Alcohol soluble extractive

Alcohol soluble extractive value of *Saraca indica* (18.33 %) along the material collected in different season does not show significant variation (Table-1) .The closest value to the substitute/adulterant was of *Polyalthia longifolia* (16.77%) while the values of *Bauhinia variegata*, *Shorea robusta* , *Trema orientalis* and *Bombax ceiba* was found to be 11.64%, 11.39%, 10.94% and 7.04% respectively .

Water soluble extractive

Water soluble extractive value of *Saraca indica* (14.15 %) along the material collected in different season does not show significant variation (Table-). The closest value to the substitute/adulterant was of *Trema orientalis* (15.07%), but the *Polyalthia longifolia* showed the maximum extraction (19.24%) while values of *Shorea robusta*, *Bombax ceiba* and *Bauhinia variegata* was found to be 12.09%, 11.12% and 8.58 % respectively.

The alcohol soluble extractive value of the *P.longifolia* was very close and water soluble extractive value was higher than to that of *S. indica* indicates that amongst all substitutes/adulterants of *S. indica* , the *P.longifolia* showed the maximum extraction of phytoconstituents which reflect that the *P.longifolia* could be chemically rich as compared to other substitutes/adulterants of *S. indica*.

Chapter 21

General Discussion

Pharmacognosy

The present study provides innumerable characters to distinguish all the original drug plants and their substitutes/adulterants. These are discussed below.

1. *Fumaria parviflora* and its substitutes/adulterants

The presence of light brown contents in cortical parenchyma, fan shaped vascular bundle and pitted fiber tracheids in root was the characteristic features of *Fumaria parviflora*. All these characters are conspicuously absent in all substitutes/adulterants. It also showed the presence of reticulate and scalariform thickened vessels. The stomata present in the leaves of *F. parviflora* was of anomocytic type and mesophyll of leaf was not differentiated into palisade and spongy parenchyma. In stem of *F. parviflora* the vascular bundle was capped with sclerenchymatous sheath. In case of *Justicia procumbens*, the centre wood dominated by vessels and vessels had elongated parallel bordered pits while in the stem the presence of interfascicular wood prosenchyma and thick-walled unicellular as well as multicellular uniseriate trichomes with broad basal cell, blunt tip and warty walls and thin and wavy lateral walls of epidermal cells were the characteristics. The leaves of *Oldenlandia corymbosa* showed the presence of paracytic type of stomata, cells of upper epidermis about double the size of the lower epidermis and network of lobed spongy cells and association of libriform fibres with tracheids in stem was characteristic but beside this the presence of raphide bundles was the striking character. The leaf of *Peristrophe bicalyculata* showed the presence of glandular and thin walled unicellular as well as multicellular uniseriate trichomes with pointed tip while the stomata was of diacytic type and the cells of epidermis were wavy. Brown deposits in cortical cells and isolated acicular crystals in pith of stem were also characteristic. *Polycarpea corymbosa* showed the many diagnostic characters such as leaves showed anomocytic type of stomata outer walls of epidermis thick and papillose, parenchyma containing reddish brown contents, vascular bundle capped by thick walled sclerenchymatous sheets and rosette crystals.

Absence of rays in root and stem was also characteristics. In case of *Rungia repens*, the stem showed the thick walled and rosette crystals while its leaves showed the presence of diacytic type of stomata and thick-walled unicellular as well as multicellular uniseriate trichomes with broad basal cell, blunt tip and warty walls here the stem showed the typical pitted sclerenchyma in a pairs wherein the adjoining walls were straight.

Among these plants trichomes were found absent in both *F. parviflora* and *O. corymbosa* while it was present in all other substitutes/adulterants viz. *P. corymbosa, J. procumbens, R. repens* and *P.bicalyculata*. Among them, *P. corymbosa* differs in having branched trichomes while *R. repens* and *J. procumnbens* differ in having multicellular uniseriate trichomes. *J. procumbens* differs in showing groups of sclereides and *P. bicalyculata* showing presence of aciaular crystals. *O. corymbosa* differs in having raphide bundles.

2. *Bergenia ligulata* and its substitutes/adulterants

The *B. ligulata* rhizome showed the distinct characters such as presence of various shaped starch grains typically having beak and rosette crystals in cork, phelloderm, cortex, ray and in pith, loosely arranged cortical parenchyma showing light brown deposits, 'V' shaped vascular bundles with cambium and spiral and simple pitted vessels. The important characters of *Aerua lanata* root were thin walled cork cells, narrow compactly arranged cortical parenchyma filled with rosette crystals and occasionally with rhomboidal crystals. The root also showed secondary anomalous secondary growth where in successive rings separated by thin walled parenchyma and separated from the central phloem by thick walled parenchyma contained rosette and rhomboidal crystals. The condition of central wood was triarch was also characteristics. The root of *Ammannia baccifera* showed the diagnostic characters such as thick walled light brown coloured cork cells, rosette crystals and presence of collenchyma between primary and secondary vascular bundles and thick walled fibres with pointed ends. The presence of thick walled brown colour cork cells, microspheroidal crystals and angular boarded pitted vessels were the characteristic of *Celosia argentea.* The root of *Coleus amboinicus* showed presence of cortical parenchyma with the reddish brown deposits, stone cells, thin and thick walled sclereids and scalariform vessel having oblique end walls while the diagnostic characters observed in root of *Glossocardia linearifolia* were cork with thick light yellow coloured wavy walled cork cells, cortical parenchyma with reddish-brown

deposition, stone cells, thick walled ray parenchyma with pits and blunt ends and broad lumen fibres.Among these plants the important diagnostic character such as presence of rosette crystals in *B. ligulata,* were also found in *Aerua lanata* and *Ammannia baccifera* while it was found absent in *Coleus amboinicus, Celosia argentea* and *Glossocardia linearifolia.* Another important character of *B. ligulata* was the presence of starch grains was also found in *C. amboinicus* but it was big, simple, various shaped and having beak and plenty in *B. ligulata* while in *C. amboinicus* it was small, simple and compound, spherical and few. The vascular bundles in *B. ligulata* was 'v' shaped, while *Aerua lanata, Ammannia baccifera* and *Celosia argentea* showed the anomalous secondary growth of vascular bundles. In case of *Aerua lanata* the vascular bundles were separated from the central phloem by thick walled parenchyma containing rosette and rhomboidal crystals while in case of *Ammannia baccifera* the primary and secondary vascular bundles separated by layers of collenchyma containing rosette crystals and *Celosia argentea* showed discrete secondary vascular bundles separated by 2 layered thick medullary rays of rectangular cells. None of the substitutes/adulterants of *B. ligulata* were showed the deposition of light brown contents in ray cells which was specific to the *B. ligulata*. The absence of pith cells in substitutes/adulterants of *B. ligulata* was also distinguishing features as the source of all substitutes/adulterants was of root. The presence of crystalline matters in *C.argentea*, rhomboidal crystals in *Aerua lanata,* thick cellulosic wall of parenchyma in *Ammannia baccifera* and stone cells in *G. linearifolia* was the distinguishing characters.

3. *Glycyrrhiza glabra* and its substitutes/adulterants

Microscopically *Abrus precatorius, Alysicarpus longifolius,* and *Taverniera cuneifolia* the substitutes/adulterants of *Glycyrrhiza glabra* were similar to *Glycyrrhiza* in characters like starch grains and prismatic crystals of calcium oxalate. *Maerua arenaria* was very different in the absence of such characters and in having parenchyma with oil droplets. The absence of tyloses and indistinct cambium in *T.cuneifolia* were the distinguishing characters and it was also differed in having characters like collapsed condition of phellogen, polygonal shaped cortical parenchyma, typical found in a pairs wherein the adjoining walls were straight while *Alysicarpus longifolius* and *Abrus precatorius* were differ in having ray parenchyma containing prismatic crystals and stone cells respectively and both had indistinct

phellogen. In *Alysicarpus longifolius* the vessel were characteristically associated with pitted parenchyma.

4. *Polygala senega* and its substitutes/adulterants

In case of substitutes/adulterants of *Polygala senega,* all substitutes/adulterants are look different. *Acalypha indica* differs in having rosette crystals, *Adhatoda vasica* in having pitted stone cells with distinct striations, *Polygala chinensis* showing deposition of pale yellow amorphous masses in some cortical and phloem parenchyma of shows parenchyma and *Xerornphis spinosa* differs in having starch grains.

5. *Saraca indica* and its substitutes/adulterants

The diagnostic characters observed in the bark of *Saraca indica* were brown thick walled cork cells, spherical starch grains, thick walled narrow lumened stone cells, sclereids, rhomboidal crystals, septet and aseptet fibers, crystal fibres.The bark of *Bauhinia variegate* showed the presence of distinct characters such as thin and thick walled cork cells, parenchyma containing reddish-orange deposits, starch grains, sclereids, thick walled narrow lumened fibers, rhomboidal and prismatic crystals. The presence of stone cells in the medullary rays and association of it with crystal fibers was also characteristics. *Bombax ceiba*, *Polyalthia longifolia*, *Shorea robusta*, and *Trema orntentails* closely resemble to *Saraca* in terms of microscopic characteristics like starch grains, crystals and stone cells. The differences observed are thin walled stone cells with simple pits in *Bauhinia variegata*, thick walled stone cells with narrow lumen in *Bornbax ceiba,* acicular crystals in *Polyalthia longifolia,* thick walled stone cells with distinct striations and pit canals in *Shorea robusta* and parenchyma with yellow content and elongated stone cells with branched lumen in *Trerna orientalis.*

Besides these characters the organoleptic characters like colour, surface taxture, fracture etc. of individual drugs mentioned in early Chapters which was characteristic to that drugs could be used for differentiating the genuine drugs from their substitutes/adulterants.

Phytochemistry

1. *Fumaria parviflora* and its substitutes/adulterants

In case of the substitutes/ adulterants of *Fumaria parviflora, Oldenlandia corymbosa* seems to close to that of *Fumaria* in having flavonol quercetin, 3'-0Me quercetin, phenolic acid vanillic acid and presence of alkaloids, steroids and tannins, while it differs in having the flavonol 3', 4'-di OMe quercetin, phenolic acids syringic, ferulic *(cis-and trans-* isomers), *p*-hydroxy benzoic, protocatechuic, melilotic acids, and iridoids. *Rungia repens* is similar in having quercetin, vanillic acid, alkaloids, steroids and tannins and differs in having 7'- OMe quercetin and kaempferol along with syringic, ferulic *(cis-* and *trans*-isomers), *p*-coumaric and p-hydroxybenzoic acids. *Polycarpea corymbosa* is similar in having vanillic acid, alkaloids, steroids and tannins and differs in having apigenin, acacetin, 3'- OMe luteolin, 7,3-di OMe quercetin. *Justicia procumbens* and *Peristrophe bicalyculata* are similar in having vanillic acid and steroids but *Justicia* differs in having 6-0H kaempferol and 7- OMe 6- OH kaempferol, and ferulic (*cis*-and *trans*- isomers) acid and *Peristrophe* in having ferulic *(cis-and trans-* isomers) and *p*-hydroxy benzoic acids.

2. *Bergenia ligulata* and its substitutes/adulterants

In case of *Bergenia ligulata, Ammannia baccifera* is similar to *Bergenia* in having steroids, vanillic, syringic and gallic acids and differs in having alkaloid ephedrine and melilotic acid. *Coleus* similar in showing presence steroids, vanillic and syringic acids and differs in having melilotic acid. *Aerua lanata* similar in having vanillic, syringic acids and differ in having quinones and ferulic *(cis-and trans-* isomers) acid whereas *Glossocardia linearifolia* and *Celosia argentea* are similar in having steroids, vanillic and syringic acids but *Glossocardia* differs in having flavonol acacetin and ferulic (*cis-* and *trans-* isomers) acid and *Celosia* differs in having phenolic acids such as *p*-coumaric and melilotic acids.

3. *Glycyrrhiza glabra* and its substitutes/adulterants

Among the substitutes/adulterants of *Glycyrrhiza, Abrus precatorius* and *Taverniera cuneifolia* seem to be more close to that of *Glycyrrhiza* in having saponin, coumarins, alkaloids, steroids and phenolic acids such as vanillic, syringic, ferulie

(cis- and trans-isomers) but, *Abrus* differs in having quinones and melilotic acid and *Taverniera cuneifolia* in having *o*-coumaric and *p*-hydroxybenzoic acids whereas *Maerua arenaria* and *Alysicarpus longifolius* are similar in having saponin, steroids, vanillic and syringic, acids while differs in having melilotic acid.

4. *Polygala senega* and its substitutes/adulterants

In case of *Polygala senega,* all substitutes/adulterants are similar to that of *senega* in having saponins, vanillic and syringic acids but *Acalypha indica* differs in having alkaloids, *p*- coumaric and ferulic(*cis*- and *trans*-isomers) acids. *Adhatoda vasica* differs in having alkaloids, *p*- coumaric and *p*-hydroxy benzoic acids, *Polygala chinensis* in having coumaric acids and *Catunaregam spinosa* in having scopoletin.

5. *Saraca indica* and its substitutes/adulterants

Among the substitutes/adulterants of *Saraca asoca, Trema orientalis* seems to be more close to that of *Saraca asoca* in showing the presence of flavonoids, steroids, cyanidin, vanillic and syringic acids but differs in having ferulic *(cis*- and *trans*-isomers)acids. *Bauhinia variegata* is similar in having steroids, vanillic and syringic acids but differs in having kaempferol and *o*-coumaric acid. *Bombax ceiba* is similar in having cyanidin, vanillic and syringic acids but differs in having ferulic *(cis*- and *trans*- isomers) acid, whereas *Polyalthia longifolia* and *Shorea robusta* are similar in having vanillic and syringic acids but *Polyalthia* differs in having ferulic *(cis*- and *trans*- isomers)acid and *Shorea* in having 3- OMe quercetin, pelargonidin derivatives and *p*- hydroxy benzoic acids.

The work on all these substitutes/adulterants provided a large amount of data useful in understanding the multifarious activities of these drugs and in quality control procedures. The phytochemical studies revealed that all the substitutes/adulterants, in spite of having more dissimilarity are rich in phytochemicals such as **flavonols, anthocyanidins, phenolic acids and mucilages.** Flavonols are the most common flavonoids. The flavonols located are kaempferol, quercetin, acacetin etc. and their various methoxylated its derivatives. Almost all these compounds are physiologically active bioflavonoids. Tannins especially proanthocyanins also are widely distributed in these plants. Alkaloids were rather rare, located in only few plants. The phenolics reported above are found to exert a multitude of pharmacological properties. Of late,

Flavonoids which are found to be the important components of the medicinal plants screened here are found to have profound health benefits. **Quercetin,** the most common flavonol**,** appeared to have many beneficial effects on human health including cardiovascular protection, anti-cancer activity, antiulcer effects, antiallergic activity, cataract prevention and antiviral and anti-inflammatory effects (Miller, 1996) and also inhibits lipid peroxidation *in vitro.* **Kaempferol** had a stimulatory effect on alkaline phosphatase activity in MG-63 human osteoblasts through ERK and estrogen receptor pathway. It was also shown to inhibit proliferation and increase mediator content in human leukemic mast cells. The activities of phenolic acids, which are found to be omnipresent, as well as those of quinones also are well recognised.

Another important chemical component overlooked in almost all the studies are the simple phenolics such as phenols, phenolic acids, coumarins etc. They are the minor components, which are never attributed with any activity. It is in this context, Duke's (1997) observations are interesting and informative. Duke (1997) describes ferulic acid, gentisic acid, kaempferol glycosides and salicylic acid as pain relievers while ascorbic acid, cinnamic acid, coumarin and resveratrol are explained to be anti-inflammatory. The present study revealed that all the plant parts screened contained a variety of phenolic acids, that too in good concentrations. These compounds may exert their own specific pharmacological actions or add to the activities of other compounds or act as efficient antioxidants. This variety of chemicals and their richness (concentration) in a medicinal herb is of great value in assessing its property. Duke's database states that both coriander and liquorice contain 20 chemicals with antibacterial action; oregano and rosemary have 19; ginger17; nutmeg15; cinnamon and cumin 11; Black pepper 19; Bay 10 and garlic 13. Quantity wise, liquorice contains up to 33%; bactericidal compounds (dry weight basis), thyme 21%, oregano 88%, rosemary 4-8%, coriander 22% and fennel 1.5%.

The role of **mucilages** in general in medicinal plants is never understood properly. It was not considered as a pharmacologically active component. But of late, the mucilages are found to exert a large number of pharmacological actions. They were known laxative agents, demulcents, emollients and anti-diarrhoeal agents. They also check fermentation, bacterial growth and adsorb toxins and wastes helping their elimination from the body. Even cholesterol is being lowered by these mechanisms, as also there are repots in which they cause a blood sugar lowering effects. Immunostimulating activities of polysaccharides have been brought out recently into

light by studies on traditional Chinese and Japanese medicines (Chang, 2002). Anti-ulcerogenic activities and stimulating proliferation of bone marrow cells are the other activities attributed to polysaccharides (Pengelly, 2006). In this context the presence of mucilages in all the drugs screened, that to in good amounts, is highly significant. These mucilages are found to be differing in their monomer composition, and also vary from plant parts to plant parts in their chemistry. The activities of these mucilages is to be studied in detail to understand their role in all these substitutes/adulterants or other medicinal plants.

HPTLC fingerprinting

Though the methods of extraction and chromatographic conditions for all the substitutes/adulterants were kept identical, the HPTLC chromatograms obtained when the HPTLC plate was scanned at 254 nm and 366 nm showed immense variations.

HPTLC profile of *F. parviflora* and its substitutes/adulterants observed under UV 254 nm showed that *J. procumbens* found more close to *F. parviflora* as it is similar in 3 peaks next is *O. corymbosa* is similar in only 2 peaks whereas under UV 366 nm *J. procumbens* is similar in 4 peaks, *R.repens* in 3 peaks, *P. corymbosa* and *P. bicalyculata* similar in 2 peaks. while *O.corymbosa* was not show any peaks similar to that of *F. parviflora*. This indicates that *J. procumbens* has more common compunds and is the closest to *F. parviflora.*

HPTLC profile of *B.ligulata* and its substitutes/adulterants observed under UV 254 nm showed that presence of 5 similar peaks keep the drug more close to *B.ligulata* while *G. linearifolia* is not show any common peak to that of *B.ligulata* showing that *G. linearifolia* is total different.

HPTLC profile of *G. glabra* and its substitutes/adulterants observed under UV 254 nm showed that *T. cuneifolia* was similar in 3 peaks while *Aberus precatorius* and *Alysicarpus longifolius* were similar in 1 peak and *Maerua arenaria* was not show any peak similar to that of *G. glabra* showed that among all substitutes/adulterants, *T. cuneifolia* is found more close to *G. glabra.*

HPTLC profile of *Polygala senega* and its substitutes/adulterants showed that at UV 254 nm *Catunaregam spinosa* is similar in 2 and under UV 366 nm 4 peaks seemed that *Catunaregam spinosa* is the closest drug to *Polygala senega*.

HPTLC profile of *S. indica* and its substitutes/adulterants observed under UV 254 nm showed that *P. longifolia* was similar in 2 peaks and *T.orientalis, B. ceiba* and *S. robusta* are similar in 1 peak only but at UV 366nm *T. orientalis* is similar in 3

peaks and *P. longifolia* in one peak only showing that *T. orientalis* is more close to *S. indica* followed by *P. Longifolia.*

However, the HPTLC fingerprint for each substitutes/adulterants needs to developed separately as a quality control parameter for the plant.

Physico-chemical analysis

Total ash content

The total ash content of a crude drug is the inorganic residue remaining after incineration. It includes not only the inorganic salts, e.g. Calcium oxalate, occurring naturally in the drug; but also inorganic matter from external sources. The ash value may be useful in to ensure the absence of an undue proportion of extraneous mineral matter introduced accidently or by design due to the type of manufacturing process used, e.g. earth, sand, floor sweepings etc. and to detect adulteration of the drug. In this study the total ash content of all the drugs does not show significant variation along the material collected in different season. Total ash content of *Fumaria parviflora* and its substitutes/adulterants along the material collected in different season does not show significant variation and the closest value to the substitute/adulterant is *Polycarpea corymbosa* while other substitutes/adulterants have higher ash values i.e almost double the value of *Fumaria parviflora*. In case of *Bergenia ligulata* the closest value to the substitute/adulterant is of *Aerua lanata* and *Coleus amboinicus* but diffrence in values are of more than 3%. Among the substitutes/adulterants *Glycyrrhiza glabra, the total ash value of Taverniera cuneifolia is almost equal to Glycyrrhiza glabra. The ash values of Adhatoda vasica* and *Polygala chinensis* are found to be close to *Polygala senega* as compare to other the substitute/adulterant while in case of substitutes/adulterants of *Saraca indica, Shorea robusta* and *Bombax ceiba* are close to *saraca indica*.

The Total Ash content mainly is a measure of the presence of inorganic compounds. A larger value indicates that the plant material contains more of inorganic compounds.

Acid insoluble ash content

Crude drugs containing larger quantity of calcium oxalate, can give variable results depending upon the conditions of ignition. Treatment of the ash with Hydrochloric acid leaves virtually only silica. Hence acid insoluble ash forms a better test to detect and limit excess of soil present as an impurity in the drug, than does the total ash. In this study the acid insoluble ash content of all the drugs does not show significant variation along the material collected in different season. In case of acid insoluble ash content of *Fumaria parviflora* the closest value to the substitutes/adulterants is of *Oldenlandia corymbosa.* Amongst other substitutes/adulterants, the acid insoluble ash content of *Coleus amboinicus* , *Celosia argentea* and *Aerua lanata* and among substitutes/adulterants of *Glycyrrhiza glabra* , the *Abrus precatorius, Taverniera cuneifolia, Alysicarpus longifolius* are found tobe more close to that of genuine drug. The acid insoluble ash content of substitutes/adulterants of *Polygala senega* does not show much variatons, whereas the acid insoluble ash content value of aduterant *Shorea robusta* is same as to that of *Saraca indica.*

Alcohol soluble extractive

These are indicative of the approximate measure of chemical constituents of crude drug. The closest value of alcohol soluble extractive to their substitutes/adulterants of *Fumaria parviflora, Bergenia ligulata, Glycyrrhiza glabra, Polygala senega* and *Saraca indica* are of *Rungia repens, Glossocardia linearifolia, Taverniera cuneifolia, Polygala chinensis* and *Polyalthia longifolia* respectively.However extractive value of *Celosia argentea* showed the maximum and is higher than that of *Bergenia ligulata.*

Water soluble extractive

Amongst all substitutes/adulterants of *Fumaria parviflora, Bergenia ligulata, Glycyrrhiza glabra, Polygala senega* and *Saraca indica* show the closest value of water soluble extractive are shown by *Justicia procumbens, Glossocardia linearifolia, Taverniera cuneifolia , Polygala chinensis* and *Trema orientalis* respectively. However extractive value of substitutes/adulterants *Rungia repens,Celosia argentea, Polygala chinensis* and *Polyalthia longifolia* showed the maximum, higher than that of their genuine drugs.

Higher values of water and alcohol soluble extractive indicate the richness of the plant material in context to its phytochemicals.

Chapter 9

Summary and Highlights

Substitution/Adulteration of herbal drugs is a rampant phenomenon at all places. Because the adulterants also share a number of characters with the genuine drugs, it becomes imperative to recognize them in the raw material, powder or extract form. Studies on these aspects are seldom conducted and for this reason, in the present work, an attempt is made to conduct a systematic study on substitutes/adulterants of five medicinally important plants widely used in Indian systems of medicine.

The parameters that can aid in rapid identification such as micromorphological studies, powder characteristics, phytochemical analysis, purity and variation in the HPTLC fingerprints were looked into during the present study. Micromorphology can be used to detect adulteration when the plant is obtained in fresh form. In case of dried plant powder, differences in powder characteristics as well as HPTLC fingerprint profiles can be utilized to ascertain the purity of the given plant powder.

The genuine drugs selected for the present study are 1) *Fumaria parviflora,* 2) *Bergenia ligulata,* 3) *Glycyrrhiza glabra,* 4) *Polygala senega,* and 5) *Saraca indica* and the substitutes/adulterants studied are *Oldenlandia corymbosa, Peristrophe bicalyculata, Polycarpea corymbosa, Justicia procumbens* and *Rungia repens* (all for *Fumaria*); *Aerua lanata, Ammannia baccifera, Celosia argentea, Coleus ambonicus* and *Glossocardia linearifolia* (all for *Bergenia*); *Taverniera cuneifolia, Abrus precatorius, Alysicarpus longifolius, Maerua arenaria* (for *Glycyrrhiza); Polygala chinensis,Acalypha indica*, *Adhatoda vasica, Catunaregam spinosa*(all for *Polygala*); and *Trema orientalis, Polyalthia longifolia*, *Bauhinia variegata*, *Bombax ceiba* and *Shorea robusta* (for *Saraca*). Standard procedures were followed for phytochemical, pharmacognostical, physicochemical and HPTLC fingerprint analysis.

The characteristic features of ***Fumaria parviflora*** which distinguished it from the substitutes/adulterants were light brown contents in cortical parenchyma and fan shaped vascular bundle in root. The diagnostic characters such as presence of raphide

bundles in *Oldenlandia corymbosa*, acicular crystals in *Peristrophe bicalyculata* and presence of trichomes in *Polycarpea corymbosa, Justicia procumbens* and *Rungia repens* which are specific to the plants are not found in *Fumaria parviflora* . All these characters could be used to distinguish *Fumaria parviflora* from their substitutes/adulterants. Chemically, syringic acidwhich is commonly found in all adulterants, is absent in *Fumaria parviflora.* The characters like presence of 3', 4'-di OMe quercetin, ferulic *(cis*-and *trans*- isomers), *p*-hydroxy benzoic, protocatechuic and melilotic acids in *Oldenlandia corymbosa*, 7'- OMe quercetin and kaempferol ,syringic, ferulic *(cis*- and *trans*-isomers), *p*-coumaric and *p*-hydroxybenzoic acids in *Rungia repens*, apigenin, acacetin, 3'- OMe luteolin and 7,3-diOMe quercetin in *Polycarpea corymbosa*, 6-0H kaempferol and 7- OMe 6- OH kaempferol, and ferulic (*cis*-and *trans*- isomers) acid in in *Justicia procumbens* and ferulic *(cis-and trans*-isomers) and *p*-hydroxy benzoic acids in *Peristrophe bicalyculata* are found absent in *Fumaria parviflora* and these could be used to distinguish all the substitutes/adulterants and their genuine drug plant. As all these compounds have various pharmacological effects, they could be considered as active principles of those plants. From among the substitutes/adulterants, *Oldenlandia corymbosa* is chemically more similar to *Fumaria parviflora* as both have same chemistry.

Similarly the characteristic features like cortical parenchyma with light brown deposits, 'V' shaped vascular bundles with cambium and various shaped starch grains typically having beak, of ***Bergenia ligulata*** were found absent in their substitutes/adulterants while rhomboidal crystals in *Aerua lanata,* presence of collenchyma between primary and secondary vascular bundles in *Ammannia baccifera*, microspheroidal crystals and angular boarded pitted vessels in *Celosia argentea* and stone cells in *Glossocardia linearifolia* which are specific to the plant used as distinguishing characters. Chemically the presence of *p*-hydroxy benzoic acid, found in high concentration in *Bergenia ligulata,* was found absent in their substitutes/adulterants this is the strong character differentiating the original drug. The phytochemical which are found absent in *Bergenia ligulata,* like ephedrine and melilotic acid in *Ammannia baccifera* and *Coleus*, ferulic *(cis-and trans*-isomers) acid in *Aerua lanata*, *p*-coumaric and melilotic acids in *Celosia*, and acacetin in *Glossocardia* are also can be used as the individual chemical characters of the substitutes.

The substitutes/adulterants of ***Glycyrrhiza glabra*** showed the more similarity except *Maerua arenaria* in having parenchyma with oil droplets and in the absence of starch grains and prismatic crystals, the characteristic of *Glycyrrhiza glabra*. The minor difference among other adulterants are the presence of tyloses in *Glycyrrhiza glabra* was found absent in all substitutes/adulterants. Chemically *Taverniera cuneifolia* shared maximum number of common compounds with *Glycyrrhiza glabra* and can be differentiated in having *o*-coumaric and *p*-hydroxybenzoic acids while *Abrus* , *Maerua* and *Alysicarpus* in having melilotic acid

In case of substitutes/adulterants of *Polygala senega,* the diagnostic character such as broad 'V'- shaped medullary rays , collenchyma with oil droplets and thick walled wood parenchyma are conspicuously absent in all substitutes/adulterants. Other major differences are presence of rosette crystals in *Acalypha indica,* stone cells in *Adhatoda vasica,* deposition of pale yellow amorphous masses in some cortical and phloem parenchyma in *Polygala chinensis* and starch grains in *Catunaregam spinosa* which are found absent in *Polygala senega.* These characters could be used for differentiating the substitutes/adulterants. They can also be differentiated on basis of presence or absence of phenolic acids such as presence of ferulic(*cis*- and *trans*-isomers) acid in *Acalypha indica, p*-hydroxy benzoic acid in *Adhatoda vasica* , coumaric acid in *Polygala chinensis* and scopoletin in *Catunaregam spinosa.* All these compound were found absent in *Polygala senega.*

Microscopically, between ***Saraca indica*** and its substitutes/adulterants there are not much differences observed, except for the presence of continues bands of stone cells in *Saraca indica* which was absent in all their substitutes/adulterants. Other characteristic features like rosette crystals in *Bombax ceiba,* acicular crystals in *Polyalthia longifolia,* gum ducts in *Shorea robusta,* and parenchyma with yellow content and elongated stone cells with branched lumen in *Trema orientalis* were found absent in *Saraca indica*. Such characters could be used as their distinguishing characters. The phytochemicals such as kaempferol and *o*-coumaric acid in *Bauhinia variegata,* ferulic *(cis*- and *trans*- isomers) acid in *Bombax ceiba*, *Polyalthia longifolia* and *Trema orientalis* and 3'- OMe quercetin and *p*- hydroxy benzoic acid in *Shorea robusta* (which found absent in *Saraca indica*) are the distinguishing characters of the plants containing them.

Ash and extractive values of all the plants taken up in this study can be used as characters of those plants.

A chromatographic fingerprint profile can establish the identity of a plant. In view of this fact, the HPTLC fingerprints of the all five drug plants and their substitutes/adulterants developed during the course of this study and compared with those of the respective adulterant plants were showing enough variations. Since all other experimental conditions were constant, the difference in the chromatographic profiles could be used to differentiate between two plants, and thus detect any adulteration in the given plant material

The present project resulted in finding out the pharmacognostic and phytochemical biomarkers and chemical diversity of all the drugs selected. The biomarkers are extremely useful in identifying the genuineness of the drug and also to find out adulteration.

Similarly ,the transverse sections of roots, leaves and stems will be of immense use in checking the identity of a medicinal plant. The powder study will help in finding out whether a powdered drug is genuine or adulterated. Locating a particular cell component, not reported from the source plant, in a powdered sample proves that the sample is adulterated. A little bit of plant debris settled at the bottom of a container having an extract will yield very valuable information on the source plant.

Highlights

The highlights of the present investigation are the following.

1. In terms of chemical constituents none of the substitutes/adulterants of five genuine drug selected are as potent as the genuine drug. However the adulterant *Oldenlandia corymbosa* chemically found to be more close to the genuine drug *Fumaria parviflora* and *Taverniera cuneifolia* to *Glycyrrhiza glabra*. All other substitutes/adulterants were found to show much differences and cannot be used as substitutes.

2. Though most of the substitutes/adulterants cannot replicate for the genuine drug, they contained biologically active compounds such as, flavonoids, phenolic acids, mucilages which exhibit a number of pharmacological properties and therefore can be used as separate drugs.

3. Flavonoids were absent in most of the roots, except for *Glossocardia linearifolia* where flavone acacetin was located. All the roots contained a good number of phenolic acids. The mucilages which were omnipresent were consisting of homopolysaccharides like galactans or heteropolysaccharides like glucomannans. The phenolic acids and the mucilages present in the root are pharmacologically active and therefore may add to the clinical properties of the drug and in many cases may act synergistically with the alkaloids in improving the drug action.

4. The most important discovery was the isolation of Ephedrine from the roots of *Ammania baccifera*. Ephedrine is used to treat breathing problems (as a bronchodilator), nasal congestion (as a decongestant), low blood pressure problems (orthostatic hypotension), or myasthenia gravis. This drug has also been used to treat certain sleep disorders (narcolepsy), menstrual problems (dysmenorrhea), or urine-control problems (incontinence or enuresis). Ephedrine, is a CNS stimulant; its immediate effects are attributable to stimulation of dopamine release. Ephedrine is defined as a mixed sympathomimetic agent that acts by enhancing the release of norepinephrine from sympathetic neurons and by stimulating alpha and beta adrenergic receptors. Ephedrine stimulates the nervous system to enhance mood, reduce fatigue, and to make a person alert enough to smell their coffee in the morning. It also has the ability to increase energy and endurance; it does this through increase of blood flow to the muscles, resulting in an increase of oxygen and nutrient supply to the muscles. Ephedrine also increases basal metabolic rate (BMR), so that the body is spurred to burn calories faster, and so ephedrine is part of the thermogenic process that can result in substantial weight loss.

5. The present study unearthed a number of new sources of flavonoids, phenolic acids and polysaccharides like mucilages.

6. Chemical and pharmacognostical markers of twenty eight drug plants such as *Fumaria parviflora, Bergenia ligulata, Glycyrrhiza glabra, Polygala senega, Saraca indica, Oldenlandia corymbosa, Peristrophe bicalyculata, Polycarpea corymbosa, Justicia procumbens* and *Rungia repens, Aerua lanata, Ammannia baccifera, Celosia argentea, Coleus ambonicus, Glossocardia linearifolia, Taverniera cuneifolia, Abrus precatorius, Alysicarpus longifolius, Maerua arenaria, Polygala chinensis,Acalypha*

indica, Adhatoda vasica, Catunaregam spinosa, Trema orientalis, Polyalthia longifolia, Bauhinia variegata, Bombax ceiba and *Shorea robusta* are developed in the present study.

7. HPTLC fingerprints of the all five drug plants and their substitutes/adulterants developed showed the enough variations.

8. The Ash and extractive values obtained in this study showed the purity and strength of that drug and extensive use in identifying the individual drugs and their quality control analysis.

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