Since the beginning of human history, plants have played an integral role in human survival. Scientists have calculated that humans in their modern form speciated only approximately 200,000 years ago, primitive vascular plants originated in the Silurian period, roughly 430 million years ago. The oldest fossils identified definitively as angiosperms date from the early Cretaceous Period, around 127 million years ago. Thus the origin of medicinal activity in plants must have been in response to environmental, not human, pressures. There is a considerable amount of evidence that herbivory was a major selective force on the evolution of plant defenses, which are typically used for medicinal applications.

The antagonistic arms race between plants and insects is evidenced by the diversification of insects and plants which occurred over the same approximate time periods, and many of these insect/host plant relationships have been conserved (Farrell 1994). The flowering plants became more diverse, so did the insects whose feeding habits were closely linked with the characteristics of the flowering plants, supporting the idea that insects and flowering plants coevolved with one another (Raven and Johnson 1989). In response to attacks by herbivores, plants have evolved a variety of mechanisms for defense and to produce a wide range of bioactive compounds. Those are broadly classified into secondary metabolites. While secondary compounds are produced in low abundance, they are essential to human and are used for various medicinal purposes (Stamp 2003).
Apart from the herbivores and insect induced response on secondary metabolites production, the plant-pathogen interaction can also cause the production of secondary metabolic compounds in many flowering plants. Structural changes such as periderm development, cellular suberization, and lignifications have been reported as the most important aspects involved in wound repair as a mechanism of defense against plant pathogens secrete and store secondary chemical compounds (Biggs 1986). Many plant pathogen interaction for instance *Fusarium roseum* f.sp. *Sambucinum*, *Cladosporium cucumerinum*, *Colletotrichum langenarium*, in various vegetables such as potatoes, yams, sweet potatoes, leaves and seedlings of cucumbers produce suberin, lignin and phenolic compounds during plant pathogen interaction (Passam *et al* 1976).

The medicinal value of secondary chemical compounds lies in its definite physiological action on the human body (Edeoga *et al* 2005). The most important bioactive constituents are such as alkaloids, tannins, flavonoids, sesquiterpenes, diterpenes, triterpene saponins, triterpene aglycones, flavonoids, sterols, coumarins, quinine, monoterpenes and phenolic compounds used for many medicinal purposes (Hill 1952). This field of natural products research is currently being carried out intensively though it remains far from exhaustion. An attempt to obtain bioactive agents from plants is a worthwhile exercise since only 10% of all plants have been investigated so far. It is imperative that ethnobotanical researches and phytochemical tests lead to some patentable and industrially exploitable compounds for drug development (Jeruto *et al* 2011). Plant derived medicines have been the first line of drug currently being used for health and combating diseases in human beings. Around 1980’s roughly
121 pharmaceutical products have been discovered based on the information obtained from the various medicinal systems. Chemical principles from natural sources have become much simpler and have contributed significantly to the development of new drugs from medicinal plants (Cox 1990).

Biologically active compounds from natural sources have always been of great interest to scientists working on infectious diseases. The Indian system of medicine has a rich source of knowledge. In India, around 20,000 medicinal plants have been recorded however; traditional communities are using only 7,000 - 7,500 plants for curing different diseases (Kamboj 2000). The medicinal plants has mentioned in various indigenous systems such as Siddha (600 plants), Ayurveda (700 plants) and Unani (700 plants). Those plants are currently very useful for drug discovery studies. The Ayurvedic concept appeared and developed between 2500 and 500 BC in India and is widely accepted and practiced throughout the world. Research to find out scientific evidence for claims of plants used for Indian Ayurvedic system of medicine has been intensified. Detailed research on the chemistry and pharmacology of products of plant origin are much essential and this may eventually lead to the discovery of medicine that can be used in the treatment of several diseases. Moreover, if these local Ayurvedic preparations are scientifically evaluated and disseminated properly, our indigenous population can be given better access to efficacious drug treatment and improved health status (Manandhar et al 1985). The resources of siddha system of medicine have been categorized into three groups: plant products (mulavargam), inorganic substances (thathuvargam), and animal products (jivavargam), which are characterized by means of taste (suvai), quality (gunam), potency (veeryam), post-digestive taste (pirivu), and specific action
(prabhavam), while Ayurveda recognizes all the drugs only by quality as the main character (Krishnamurthy and Mouli 1984).

Siddha system used animal products such as human and canine skulls in the preparation of special “ash” (chunnam) which is said to be effective against mental disorders. The alchemy in Siddha System of Medicine (SSM) has been found well developed into a science and highly used in medicine. The siddhars were even polypharmacists who were engaged in several alchemical operations which involved several processes such as calcinations, sublimation, distillation, fusion, fermentation, separation, exaltation, purification, extraction, incineration of metals and liquefaction. This was found useful in the preparation of medicine as well as in transmutation of basic metals into gold. Therefore, alchemy is one of the highly distinguished features of SSM when compared with Ayurveda (Wujastyk 1995).

In siddha system of medicine many plant resources have been recognized as main components in medicine preparation among which 108 herbs called as karpa mooligaigal are dominantly used in SSM for human ailments including respiratory diseases. This karpa mooligaigal consists of kayakalpam (kaya-body, mind and psyche, and kalpam – transmutation) plants. Some of the plants such as Acalypha indica, Aloe barbadensis, Azadirachta indica, Ocimum sanctum, Phyllanthus amarus, Phyllanthus emblica, Withania somnifera, Zinger officinale, Cynodon dactylon, Solanum trilobatum and Cuminum cyminum have been often present in various drugs formation (Arjune ram et al 2009). These plants are believed to transform health and consciousness to prevent and give relief even from chronic diseases. In addition, many herbal formulations are also used in this system. Even nowadays, in India some of these herbal formulations
are manufactured by pharmaceutical companies like TAMPCOL, IMCOPS, SKM SIDDHA, for their commercial use by Siddha physicians.

The Unani System of Medicine (USM) pioneered in Greece and was developed by Arabs into an elaborate medical science based on the frame work of the teaching of Buqrat (Hippocrates) and Jalinoos (Galen). The World Health Organization (WHO) has recognized the USM as an alternative system to cater the health care needs of human population. Alternative medicine is being practiced worldwide. Unani is one of the most well known traditional medicine systems and draws on the ancient traditional systems of medicine of China, Egypt, India, Iraq, Persia and Syria. It is also called Arab medicine. Unani is still popular in many Arab and East Asian countries. In fact Unani medicine and herbal products are gradually more being used in many countries where modern medicine is easily available.

Traditional medicine, also known as complementary medicine or alternative medicine, even in the present age, provides the first line of primary health-care to major segments of the population throughout the world. Traditional medicine can offer several advantages over modern medicine, otherwise known as allopathic medicine. First, since traditional medicinal system has been practiced in various countries from time immemorial and still persists in the modern age, the people have both found it as well as believe that the system is efficacious in curing diseases. Second, a holistic way of treatment is the basis of all traditional medicinal systems throughout the world; as such, traditional medicine aims at curing the whole body instead of specific symptoms, which is often done in allopathic medicine. Third, it offers a cheaper alternative; since the traditional medicinal practitioner mostly uses simple decoctions of
plants, animals or minerals, the medicine can be afforded by the poorer segments of the population, particularly in the developing countries. Fourth, since traditional medicinal practitioners are usually found in rural areas, people have better access to them than modern medical practitioners, who tend to converge in the bigger cities, and so are inaccessible to the rural population lacking road and other communication facilities. Fifth, the close proximity of the traditional practitioners with the rural population generates in the rural people, particularly women, to disclose their illnesses to the traditional practitioners; such rural women may feel hesitancy to discuss their illnesses or symptoms with a city doctor, who to her, is an alien being. Sixth, allopathic medicine cannot provide any cure for common diseases like rheumatism or diabetes but merely treats and alleviates the symptoms, while traditional medicinal practitioners claim to have complete cure of these diseases in many parts of the world. And finally, the seventh advantage is that allopathic drugs are now increasingly associated with severe side effects, and development of drug-resistant vectors. All these factors have contributed to a resurgence of interest in traditional medicine traditional medicines have a history of long usage, which sometimes go back even thousands of years ago (Atsana Khatun et al 2011)

Obviously, to serve this long period of time, the natural medicine has to be efficacious or otherwise would have been rejected by the people. That traditional medicine is still considered of value by the people has been shown by an estimate that about 64% of the total global population still remains dependent on traditional medicine for their healthcare needs (Cotton1996). Also historical evidence clearly demonstrates that a number of modern drugs have been discovered through observation of medicinal practices of indigenous peoples. Modern drugs like aspirin, atropine, ephedrine, digoxin, morphine, quinine,
reserpine and tubocurarine are examples, which were originally discovered through observations of traditional cure methods of indigenous peoples Gilani and (Rahman 2005). The same is true of Malaysian medicinal plants. Some examples of clinically useful drugs obtained from Malaysian medicinal plants include bromelain from *Ananas comosus* (L.) Merr. (Bromeliaceae), bergenin from *Ardisia japonica* Bl. (Myrsinaceae), arecoline from *Areca catechu* L. (Palmae), chymopapain from *Carica papaya* L. (Caricaceae), and ouabain from *Strophantus pratus* Baill. (Apocynaceae) (Jamal 2006). Under the circumstances, it is important to scientifically validate the use of a number of traditional medicinal plants by using the developments made in fields of Phytochemistry, phyto-pharmacology, phyto-medicine and phyto-therapy during the last decade. The ban on the use of antibiotics and other chemicals in livestock feeds since 2006 by the EU, because of the risk to humans of chemical residues in food and of antibiotic resistance being passed on to human pathogens, has further provided momentum to the research efforts on exploiting plants. Asian traditional medicinal systems such as Traditional Chinese Medicine (TCM), Korean Chinese Medicine, Japanese Chinese Medicine (kampo), Mongolian Traditional Medicine, Tibetan Medicine etc. and the integrative medicine – the combination of traditional medicine with conventional medicine could provide novel medicines for treatment of both animals and human disease.

The approach to new drugs through natural products has proved to be the single most successful strategy for the discovery of new drugs, but in recent years its use has been deemphasized by many pharmaceutical companies in favor of approaches based on combinatorial chemistry and genomics, among others. Again with rapid industrialization of the plant and the loss of ethnic culture and customs, some of the information on ethnomedicine has been lost.
An abundance of ethnomedical information on plant uses can be found in scientific literature but has not yet been compiled into a usable form. Collection of ethnomedical information especially in the developing countries remains primarily an academic endeavour of little interest to most industrial groups. The past successes of the natural products approach and also explore some of the reasons why it has fallen out of favor among major pharmaceutical companies and the challenges in drug discovery from natural products especially higher plants has to be analysed.

Secondary metabolites are extraordinarily diverse, with more than 105 structures reported to date, and they are also synthesized at a considerable rate, with approximately 20% of the carbon fixed by photosynthesis dedicated to their biosynthesis (Ververidis et al 2007). Flavonoids represent one of the largest groups of secondary metabolites, with more than 8000 different compounds described in the literature. Flavonoids are not synthesized in animal cells, thus their detection in animal tissues is indicative of plant ingestion. In the majority of flavonoids (flavanols, anthocyanidins, flavanones, flavones, and flavonols) the B-ring is attached at the 2\textsuperscript{nd} position, with different subclasses distinguished by the degrees of saturation and oxidation of the C-ring. The relatively uncommon isoflavones are a notable exception to this pattern, where the B-ring is instead attached at the 3\textsuperscript{rd} position. Each flavonoid subclass comprises numerous members, differing in the degree of hydroxylation or methoxylation of A and B rings. Additionally, various glycosylation patterns further increase the potential number of flavonoids. Flavonoids are mainly divided into seven major groups such as Flavonols, Flavones, Flavanones, Flavanols, Catechins (Proanthocyanidins), Isoflavones and Anthocyanins. One of the best described flavonoid, Quercetin is a member of this group and used for many medicinal
purpose (De Groot and Rauen 1998). In plant cells, flavonoids occur mostly as glycosides, reflecting a biological strategy apparently aimed at increasing their water solubility, at specifying their subcellular localization and, most likely, at decreasing their propensity to interact with macromolecules. Epidemiological, clinical and experimental studies have indicated dietary intake of flavonoids confers protection against multiple chronic diseases negatively impacting human health (Yao et al 2004). Globally, cardiovascular disease is the leading cause of death (WHO 2008) and the impact of flavonoid consumption on cardiovascular health has been shown in a number of studies, though more work is required to extend such a conclusion to all classes of flavonoid.

Now a days there is a great decrease in plant resources due to human disturbances of the natural environment. Therefore, biotechnologists hope for a bypass to overcome this difficulty by introducing plant tissue culture technique and further multiplication of important plants by micropropagation technique. Now, the technique of tissue culture has been developed for large-scale cultivation of plant cell. The production of useful metabolites from plant tissue culture has created a new methodology for their commercialization. The useful metabolites from plant tissue cultures include alkaloids, antimicrobials, essential oils, flavonoids, pigments, proteins, phenols, pyrethrins, rotenoids, sterols and steroids. Several products are accumulated in cultured cells at a higher level than those in native plants e.g. shikonin by *Lithospermum erythrorhizon* and diosgenin by *Dioscorea*. For more than 30 years many researchers have been investigating plant cell cultures for the production of a variety of phytochemical. A numbers of firms in US, Japan, Canada and Europe have been investigating intensively the production of a very promising anti-tumour compound, Taxol using the cell cultures (Renu Sarin 2005).
The natural habitats for medicinal plants are disappearing fast and together with environmental and geopolitical instabilities; it is increasingly difficult to acquire plant-derived compounds. This has prompted industries, as well as scientists to consider the possibilities of investigation into cell cultures as an alternative supply for the production of plant pharmaceuticals. Advances in biotechnology particularly methods for culturing plant cell cultures, should provide new means for the commercial processing of even rare plants and the chemicals they provide. These new technologies will extend and enhance the usefulness of plants as renewable resources of valuable chemicals. There has been considerable interest in plant cell cultures as a potential alternative to traditional agriculture for the industrial production of secondary metabolites (Dicosmo and Misawa, 1995). Plant cell culture technologies were introduced at the end of 1960s as a possible tool for both studying and producing plant secondary metabolites.

Studies on the production of plant metabolites by callus and cell suspension cultures have been carried out on an increasing scale since the end of the 1980's. The prospect of using such culturing techniques is for obtaining secondary metabolites, such as active compounds for pharmaceuticals and cosmetics, hormones, enzymes, proteins, antigens, food additives and natural pesticides from the harvest of the cultured cells or tissues. Biotechnological cultivation of plant cells and tissues involve two major methodologies namely, cell culture studies and clonal propagation techniques. Cell culture studies begin with callus initiation using in vitro cultures for the purpose of determining the medium that best adapts for cultivation. When calli are obtained, they can undergo somaclonal variation, usually during several subcultures (Gurel 1989).
When genetic stability is reached, callus lines need to be screened in order to evaluate the productivity of each cell line so that the best performing lines can be taken to cell suspensions. Various approaches can be used to increase the production of secondary metabolites in cell suspensions but elicitation is usually one of the most successful. The final step is the bioreactor studies leading to a possible commercial production of secondary metabolites. This is a critical step as various problems can arise when scaling-up from shake flasks to bioreactors. However bioreactors act as a biological factory indeed the production of secondary metabolites with many advantages listed as follows: controlled supply independent of plant availability, increased working volumes, homogeneous culture due to mechanical or pneumatic stirring mechanism, better control of cultural and physical environment, therefore easy optimization of growth parameters such as pH, nutrient media, temperature, etc. for achieving metabolite production, reproducible yields of end product under controlled growth conditions, enhanced nutrient uptake stimulating multiplication rates and yielding a higher concentration of yield of bioactive compounds, simpler and faster harvest of cells, the opportunity to perform biosynthetic and/or biotransformation experiments related to metabolite production with enzyme availability, easier separation of target compounds because of lower complexity of extract and better control for scale-up. Genetic engineering of a secondary metabolic pathway aims to either increase or decrease the quantity of a certain compound or group of compounds (Dixon 2001, Verpoorte and Alfermann 2000). To decrease the production of certain unwanted compound(s) can be achieved by using several approaches the gene modification studies provide an excellent option to synthesis secondary metabolites through in vitro. The best studied pathway at the genetic level is the one leading to the formation of flavonoids and anthocyanins (Forkmann and Martens 2001). Most of the genes
in the anthocyanin pathway have been cloned by a combination of biochemistry, molecular biology and genetics. Other secondary metabolite pathways have extensively been studied at the level of intermediates and enzymes mainly lead to pharmaceutically important products such as indole and isoquinoline alkaloids (Facchini 2001).

The biggest challenge of producing secondary metabolites from plant cell suspension cultures is that secondary metabolites are usually produced either by specialized cells or in specific developmental stages (Balandrin et al 1985). Some compounds are not synthesized, if the cells remain undifferentiated (Berlin et al 1985). Therefore, undifferentiated plant cell cultures often lose, partially or totally, their biosynthetic ability to accumulate secondary products (Rokem and Goldberg 1985, Charlwood and Charlwood, 1991). Although there are some reports of co-cultured differentiated tissues (e.g. shoots+roots) being used to produce secondary metabolites (Subroto et al 1996, Mahagamasekera and Doran 1998) most efforts focus on transformed (hairy) roots, the result of genetic transformation by Agrobacterium rhizogenes have attractive properties for secondary metabolite production. They often grow as fast as or faster than plant cell cultures (Charlwood and Charlwood 1991, Flores et al 1999) and do not require hormones in the medium. The greatest advantage of hairy roots is that hairy root cultures often exhibit about the same or greater biosynthetic capacity for secondary metabolite production compared to their mother plants (Banerjee et al 1998, Kittipongpatana et al 1998).

Many valuable secondary metabolites are synthesized in roots in vivo, and often synthesis is linked to root differentiation (Robins et al 1991, Flores et al 1999). Even in cases where secondary metabolites accumulate only in the aerial
part of an intact plant, hairy root cultures have been shown to accumulate the metabolites. Effort has been ongoing in the production of solasodine using cell and lately hairy root cultures of various Solanum species. As seen in the experiments, some of the hairy root lines of Solanum khasianum Clarke show enhancement of solasodine production compared to non-transformed roots (Asha Jacob and Nutan Malpathak 2004). Currently more than 2000 plant species have been intensively investigated for the isolation of bioactive compounds predominantly from the Euphorbiaceae, Asteraceae, Labiatae, Fabaceae, Meliaceae, Malvaceae, Solanaceae, Rutaceae, Polygalaceae, Apocynaceae, Papaveraceae and Apiaceae (Garcia et al 2004). Among the metabolites such as flavonoids, terpenoids, alkaloids, steroids and phenols (Orozco et al 2006) have showed pronounced activity against many diseases in test animals.

Malvaceae comprises 88 genera and 2300 species which are distributed throughout the world. The members of this family are mostly annual to perennial herbs to shrubs or small trees. The leaves, roots and stems of Abutilon indicum contain considerable amount of mucilage due to which it has been used in indigenous medicine for the treatment of rheumatism and as demulcents, emollients and diuretics. Different parts of Abutilon indicum are known to have medicinal properties for instance, flowers are used to heal the boils and ulcers, leaves were traditionally used to treat bronchitis, gonorrhea and as a mouth wash in toothache. Local practitioners have claimed that the leaves are highly useful in controlling diabetes mellitus. Barks are used as diuretic, anthelmintic, pulmonary sedative and in fever and haematurea. Root oil is reported to have analgesic activity comparable with that of acetyl salicylic acid and it is devoid of Central Nerve System depressant activity. Seeds are used as laxative in piles and in the
treatment of cough. The multifold use of this plant has created wide interest in their phytochemistry.

Chemical investigations on *Abutilon* species were undertaken as early as the 1929; when Kosakal reported the presence of anthocyanins in *Abutilon avecenae*, however real chemical research on *Abutilon* species began only in 1956 with the isolation of Rutin from *Abutilon avicenae*. Developments after 1960 progressed more rapidly mainly because of the invention of more refined techniques of separation and structure elucidation. Since then large variety of compounds have been isolated from genus *Abutilon* and majority of them are flavonoids and terpenoids such as Raffinos, Gossypetin-8-glucoside, Cyanidin-3-Rutinoside, β-sitostero (*A. indicum*), β-sitostero (*A. ramosum*), Kaempferol-3-o-β-glucopyranoside (*A. grandifolium*), Myricetin-3-o-β-Glucopyranoside (*A. theophrasti*) (Leonard 2004).

The Solanaceae family comprises nearly 102 genera and 2460 species out of which 60 to 70% of the species produce alkaloids, which play an important role against pathogens and herbivores. *Solanum surattense* is reported to contain glucoalkaloids (solasodin, diosgenin and apigenin), fatty acids, resins and mucilages. The literature survey reveals that various parts of *Solanum surattense* have been used as a folklore medicine for curing various ailments like asthma and cough (root), rheumatism (leaf), sore throat (fruit), anthelmintic (fruit), as a culminative and dropsy (plant) for relief in burning sensation in the feet accompanied by vesicular watery eruptions (plant) (Gupta and Dutt, 2009).
In light of the above mentioned facts the present study on the *in vivo* and *in vitro* secondary metabolites production in *Abutilon* and *Solanum* species was carried out with the following objectives.

- To understand the distribution pattern and localize the medicinally useful compounds in various tissues by histochemical methods.
- To standardize a reproducible regeneration protocol through callus induction, indirect organogenesis and direct organogenesis.
- To enhance the secondary metabolite (quercetin) production through *in vitro* culture studies.
- To screen the preliminary antimicrobial activity.
- To compare drug value of bioactive compounds derived from *in vivo* and *in vitro* plant source by using test animals.