CHAPTER-11
DISCUSSION

India has rich storehouse of traditional knowledge of medicinal plants. Naturally derived phytochemicals have scientifically well established therapeutic properties like antibacterial, antifungal, antioxidant, anticancer, antidiabetic and so on. In the recent years increase in the case of antibiotic resistant disease causing micro organisms and the need to develop cost effective drugs further strengthens the pressing need to explore phytochemicals from medicinal plants. Higher plants have been shown to be a potential source for the identification of new antimicrobial agents. The screening of plant extracts has been of great interest to scientists for the discovery of new drugs effective in the treatment of several diseases (Alim et al., 2009). A number of reports concerning the antibacterial screening of plant extracts of medicinal plants are available, but the vast majority has not been adequately evaluated (Sokmen et al., 1999).

In India, Cassia species is widespread; it grows from Himalayas to down south Kanyakumari. In Tamil Nadu, most of the waste lands are inhabited by one or the other forms of Cassia species. They are grown as an ornamental trees or as hedge plants due to their brilliant yellow coloured flowers. They have a place in the home remedies and traditional system of medicine. Though several reports are available, still the full potential of the plant has not been exploited, therefore the present work was carried out to
elucidate some of the biological properties of *Cassia* species. The five plants selected were *C. occidentalis*, *C. auriculata*, *C. alata*, *C. fistula* and *C. roxburgii*. Proper identification of the plant is necessary, therefore thorough morphological and anatomical features of the five plants were studied.

Angiosperm flowers are regarded as complex and integrated systems in which floral traits are organized to ensure and maximize reproduction (Chouteau, 2006). The flower morphological character of the five *Cassia* species namely *Cassia occidentalis*, *Cassia auriculata*, *C. alata*, *Cassia fistula* and *Cassia roxburghii* resulted to have almost the same floral descriptions. The result of floral morphology does not vary in individuals of same species in all areas of *Cassia fistula* and *C. alata*. This is because reproductive organs are stable which means that they are not easily affected by the environment. However, some differences in floral morphology of *Cassia fistula* and *Cassia alata* are observed, thus separating them into 2 species.

The type of inflorescence in *Cassia fistula* is a pendulous raceme while in *C. alata* the type of inflorescence is a spike. Their flowers of all five species are complete because the four major floral parts are present in its flower such as the calyx, corolla, androecium, and gynoecium. Their flowers are also bisexual because both the gynoecium and androecium are present in the same flower. Furthermore, their flowers are polycarpic because they bore flowers many times in life which would also fruit many times in life as observed in its flowering pattern. All the species have zygomorphic flowers. According to Neal *et al.* (1998) floral symmetry played an important role in the
pollination of flowering plant. The internodal elongation in flowers of both species are cardophore, this is the term that denotes a flower that has elongation of their thalamus beyond the carpel. In the type of calyx both species are gamosepalous. The type of corolla in both \textit{C. fistula} and \textit{C. alata} is described as asymmetrical and polypetalous. The development on this kind of petal depends not only on the proliferation of cells but also on the specification of petal identity.

In our observation \textit{C. occidentalis} plant is a variable, branching, erect shrub. Leaves pinnate pubescent, leaflets pale green to bluish green, 3 to 9 pairs, opposite, margin entire, flowers brilliant yellow, in erect, terminal racemes. \textit{C. auriculata} is a much branched shrub with smooth cinnamon brown bark with bright yellow, large flowers. The racemes are few-flowered, crowded in axils of upper leaves so as to form a large terminal inflorescence.

\textit{C. alata} a large shrub with very large once-compound leaves. The very large leaflets have entire margins and rounded tips. Its golden yellow flowers are borne in dense elongated clusters interspersed with yellow or orange floral bracts. Its elongated pods are somewhat four-angled and have papery wings.

\textit{Cassia fistula} is a tree with many spreading branches. The wood is hard and heavy. The flowers are large, fragrant, bright-yellow, and borne on long, slender, smooth pedicels. The racemes are axillary, pendulous, hanging down. In flowering season, the whole tree is with full blooms with few or no
leaves and the tree appears like a yellow shower. Most of the *Cassia* species produce bright yellow flowers, but *C. roxburghii* produces pink coloured flowers in small clusters.

*C. roxburghii* is a fairly large “shower” tree grows to a height of five meters with feather like pinnately compound leaves and twigs covered with a dense carpet of fine, soft hairs. Red cassia produces clusters of pink, rose or orange flowers in axillary and terminal, often branched, racemes. Ovary is superior, unilocular, with marginal ovules in all the five species. The fruit is a typical legume, it is cylindrical and indehiscent and bears many seeds separated by papery partitions.

Mohamed *et al.*, [2011] described the plant *C. occidentalis* fruit as a pod, green in color, turns into brown with light brown edges when mature, linear, distinctly compressed, straight or curved slightly upwards.

In *C. auriculata* the epidermal layer of the midrib on the adaxial side consists of circular or squarish thick walled cells which were prominent. The abaxial epidermis of the midrib consists of semicircular papillate thick walled cells. The ground cells of the midrib in the abaxial part includes 4 or 5 layers of compact parenchyma cells, on the abaxial part the palisade tissue is horizontally transcurrent extending in between the adaxial epidermis and vascular bundle.

The transverse section of the leaf let exhibits prominent midrib and thin lamina. The midrib is flat on the abaxial side and the adaxial part of the midrib consists of semi circular part with thick finger like lopsided appendage.
The midrib consists of single layer of epidermis, narrow ground tissue and bowl shaped wide abaxial vascular strand and short segment of adaxial vascular strand. The epidermal layer of the adaxial side consists of tubular, thick walled cells with smooth surface in *C.alata*.

In *C.fistula* the midrib is flat on the adaxial side and somewhat triangular on the abaxial part. The epidermis of the midrib consists of small prominently papillate thick walled epidermal cells. The vascular system consists of wide and deep main vascular strand and a small group of adaxial vascular strand. The main strand consists of several, parallel longitudinal lines of xylem elements alternating with thick walled parenchymatous cells. On the lower border of the xylem strand occur a thick discontinuous layer of phloem elements. The adaxial strand includes a few xylem elements and a large mass of phloem elements. When viewed under polarized light large prismatic calcium oxalate crystals are seen located in the parenchyma cells. The entire vascular system is ensheathed by 3 or 4 layers of thick walled lignified sclerenchyma cells.

In *C.roxburghii* the vascular strand includes fairly thick horizontal segment of xylem elements which are in short parallel lines. Beneath the xylem strand occurs a thick arc of phloem elements. The vascular strand is borne on a thick arc of sclerenchyma cells on the abaxial side and a small mass of sclerenchyma cells at top of the xylem elements. Small prismatic crystals are located in the cells present along the lower end of the sclerenchyma arc situated beneath the vascular bundle.
The stomata are paracytic type in all the five species. There are two unequal subsidiary cells occurring parallel to the long axis of guard cells.

Long, unbranched, unicellular foliar sclereids are seen in the lamina. They are mostly associated with the veins. The sclereids are seen attached with the vein and the sclereid runs along the venation. Epidermal trichomes are occasionally seen on the epidermis. The epidermal cell from which the trichome arises is circular, thick walled and darkly stained. From the circular cell originate several radiating arms of cells and these cells are celled rosette cells. These rosette cells are found in *C. auriculata*, *C. alata*, *C. fistula* and *C. roxburghii* and not found in *C. occidentalis*.

In *C. auriculata* the vein terminations occur in some of the islets and the terminations may be absent in other islets. The terminations are mostly simple, long and curved with dilated rays. There are also terminations which are forked at the tip. Foliar sclereids are commonly seen associated with the veins. The sclereids may be long and filamentous running parallel to and attached with the veins. There are also short, rectangular or slightly lobed sclereids which are attached with the veins. Epidermal trichomes are often seen in the surface view of the lamina. The trichomes are unicellular, unbranched, straight and pointed at the tip. The vein terminations are curved and slightly wavy. Running along the veins there are foliar sclereids. These sclereids are long thin and appear wavy in outline. They have thick walls and wide lumen in *C. alata*. 
Vein terminations are seen almost in all islets. The terminations vary from simple and unbranched type to repeatedly branched dendroid type in *C.fistula*. The terminations are thin and straight. Minute granular crystals are seen all over the venation system. The crystals are firmly adhering to the veins.

In *C.roxburghii* showed the terminations are short or long, thick, straight and most of them are unbranched. Foliar sclereids are invariably seen associated with the veins. The sclereids are short, rectangular or lobed cells with thick secondary walls and wide lumen. The sclereids are lying parallel to the veins or project into the vein islets.

The pollen chambers are wide and includes 4 chambers in each anther possessing scattered pollen grains in *C.occidentalis*. In *C.auriculata* the anther wall is very thick and consists of outer epidermal layer of thick rectangular cells with prominent cuticle. The pollen grains are dark brown in unstained condition. The individual pollen grains are three angled with reticulate thickenings of the exine.

In *C. alata* the anther is dithecous and two chambered. The anther wall consists of outer thin walled epidermal cells and inner thick endothecial layer which consists of radially oblong cells with highly thick and lignified radial walls. The pollen grains are triangular in outline and the exine exhibits reticulate surface thickening.

*C.fistula* shows dithecous anther and consists of two triangular lobes, each lobe having two pollen chambers. The anther lobes are wide with thick anther walls which includes on outer layer of narrow rectangular cells and inner
layer of thick walled wide rectangular cells. In between the epidermal layer occurs a row of wide vertically oblong cells with thick and lignified partition walls.

The ovary is monocarpellary and one chambered. The ovary wall is very thick and it consists of thin glabrous inner epidermal layer and densely tomentose outer epidermal layer. The embryo occurs within wide central cavity surrounded a thick cylinder of cellular endosperm. The ovary wall has well developed vascular strands. In *C.alata* ovary is dumb bell shaped with thin middle part and dilated ends.

In *C.fistula* ovary is spindle shaped with wide shallow concavity in the middle part. The ovule is thick and elliptical in shape and it includes a bulbous embryo in *C.roxburghii*. Nectariferous gland is seen in the axils of the sepal of *C.roxburghii* flower. The gland consists of thin long stalk and oblong elliptical body.

The plants were analyzed for the presence of phytochemicals in leaves and flowers separately. The leaves and flowers of all the five species were collected shade dried, powdered and ground to a fine powder. The powder was extracted with three solvents namely methanol, chloroform and petroleum ether, and the extract was evaporated and estimated for various phytochemicals and the results showed that they are rich in secondary metabolites. Phytochemical analysis of the crude extracts revealed the presence of an array of active chemical constituents such as tannins, flavonoids, glycosides,
carbohydrates, steroids and triterpenoids among which tannins play an important role.

The result of phytochemicals in the present investigation showed that the plant contain more or less same components like saponin, triterpenoids, steriods, glycosides, anthraquinone, flavonoids, proteins and amino acids. Results shows, plant rich in tannin and phenolic compounds have been shown to posses antimicrobial activities against a number of microorganisms. The present study justifies the claimed use of flowers in the traditional system of medicine to treat various infectious disease caused by the microbes. Therefore, It may be concluded from the results, that the crude extracts obtained from the flowers of Cassia species may be used enough as drug to treat disease caused by those bacteria. But before use in human being isolation of pure compound, toxicological study and clinical trial in animal model should be carried out thereafter. However, further studies are needed to better evaluate the potential effectiveness of the crude extracts as antimicrobial agents.

In present study the alkaloids and flavonoids were present in leaves and flowers of methanol extract of C.auriculata. But alkaloids were absent in other solvents of leaves and flowers. Formation of foam indicates the presence of saponins in leaves and flowers of chloroform and petroleum ether extract. Steroids, phlobatannins and fats were absent in all solvents. Thiols and amino acids were present in leaves and flower. Glycosides and tannins were present in leaves and flowers of methanol and petroleum ether.
In *C. alata* alkaloids and flavonoids were present in leaves and flowers and amino acid were present in 3 solvents. Phenols and glycosides were present in leaves of methanol and chloroform, absent in flowers. Tannins were present in leaves of methanol and chloroform extract and also present in methanol and petroleum ether extract. The preliminary phytochemical investigation were compared within *Cassia* species, and *C. roxburghii* shows the maximum presence of secondary metabolites in flowers. Minimum secondary metabolites were present in leaves and flowers of *C. occidentalis*.

Tannins are polyphenolic compounds and shown to produce anthelmintic activity [Niezen *et al.*, 1995]. Some synthetic phenolic anthelmintics like niclosamide, oxyclozanide and bithionol are shown to interfere with energy generation in helminth parasites by uncoupling oxidative phosphorylation. It is possible that tannins contained in the extracts of *Cassia auriculata* produced similar effects. Another possible anthelmintic effect of tannins is that they can bind to free proteins in the gastrointestinal tract of host animal [Athanasiadou *et al.*, 2001] or glycoprotein on the cuticle of the parasite [Thompson and Geary, 1995] and cause death.

Nayan *et al.*, [2011] studied the presence of different phytochemicals with biological activity that can be of valuable therapeutic index. Phytochemicals such as alkaloids, flavonoids, carbohydrates, glycosides, protein and amino acids, saponins and triterpenoids revealed the presence of most of constitutes in polar extracts such as ethanol, methanol and aqueous extracts compared to non polar extracts (petroleum ether and chloroform).
However, flavonoids, protein and amino acid, tannin and phenolic compounds were found to be universally occurring in all the extracts [Panda, et al., 2015; Sule, et al., 2011].

*Cassia fistula* known as Indian laburnum is a medicinal plant of immense importance. The stem bark is laxative, anti tubercular, anthelmintic, emetic, febrifuge, diuretic, depurative and useful in treatment of boils and pustules, leprosy, ring worm, colic, dyspepsia, constipation, fever, diabetic, and cardiac problems (Kirthikar and Basu, 1998; 2006). In Cambodia, the bark is used in dysentery (Prajapathi et al., 2003). The stem bark is effective in suppressing blood glucose levels and in prevention and management of coronary artery disease (Nirmala et al., 2003). It has antioxidant activity, inhibition of peroxidation, and DPPH radical scavenging ability (Sidduraju et al., 2002). Fallen cow and buffalo hides are tanned by East India tanning process using stem bark (Parthasarathy and Kamath, 1974).

The antifungal activity of different solvents was evaluated using well diffusion method. The extracts were screened for activity against *E. floccosum, M.gypseum* and *T. mentagrophytes*. Results were compared with the drug nystatin. Methanolic extract at concentration of 100 μl showed excellent zone of activity against organisms which was more or less equal to that of the standard. No appreciable zone of inhibition was observed with other solvents. The antimicrobial analysis showed a remarkable activity against the bacterial and fungal pathogens in methanol extract.
The antibacterial activity of different solvents was evaluated. The extracts were screened for activity against *E. coli*, *S.aureus* and *Vibrio* by well diffusion method. Results were compared with the drug Ciprofloxacin. Methanolic extract at concentration of 100 μl showed appreciable zone of activity against *E.coli* and *S typhi* and vibrio which was more or less equal to that of the standard. No appreciable zone of inhibition was observed with other solvents.

In our study the antibacterial activity of leaves of methanolic and chloroform extract showed appreciable zone of activity against *E.coli*. No inhibition was observed with petroleum ether. The methanol and chloroform extract of flower at concentration of 100 μl and 75 μl showed appreciable zone of activity against *E. coli* and *S.aureus*. In fungal activity the leaves of methanol and chloroform extract at concentration of 100 μl and 75 μl showed good zone of inhibition against *M.gypseum* and *T.mentagrophyte*. The methanol extracts showed significant activity against *E.floccosum* and *M.gypseum*. No sufficient zone of inhibition was observed with other solvent. In fungal activity the methanol extract of leaves at concentration of 100 μl showed excellent zone of inhibition against *E.floccosum, M.gypseum* and *T.mentagrophyte* . The flowers of methanol extracts showed significant activity against *E.floccosum M.gypseum* and *T.mentagrophyte*. No inhibition was observed with petroleum ether.

The results of the present study agrees with the findings of other workers (Gasquet, 1993; Percez, 1994; Saraf, 1994) that the leaves exhibited
in-vitro antibacterial, antimalarial and antihepatotoxic properties. The plant may be used for the treatment of colibacillosis caused by E. coli which occurs in all species of newborn farm animals as major cause of death and economic loss in this age group (Radostits et al., 2000). (Leeflang 1993) had earlier observed that indigenous knowledge and practices will be useful in the promotion of animal health and meat production in the near future in Nigeria.

Vedpriya et al., (2010) observed among all tested extracts, methanol and water extracts were found to be most active than corresponding organic extracts. Methanol extract was found to be active against six tested bacteria (P. aeruginosa, K. pneumoniae, P. mirabilis, E. coli, S. aureus, S. epidermidis). On the other hand, the aqueous extract was effective against three out of seven tested bacteria (P. vulgaris, K. pneumoniae and P. aeruginosa) and fungus (C. albicans). Aqueous extract was found to have maximum zone of inhibition against P. aeruginosa while the minimum zone of inhibition was less against K. pneumoniae. The benzene and petroleum ether extracts of the leaves of C. occidentalis were effective against P. mirabilis and E. coli respectively while chloroform extract was found to be very inactive against all tested bacterial and fungal and yeast strains. In our studies C.occidentalis showed that the methanolic and chloroform extract at 100 μl showed appreciable zone of activity against bacterial and fungal strains such as E. coli, S.aureus and Vibrio E. floccosum, M. gypseum and T. mentagrophytes.

Awal et al., (2010) reported that crude ethanol extracts of the leaf and root parts of Cassia fistula were tested against 5 Gram-positive and 9 Gram-
negative bacteria at concentrations of 30 μg and 200 μg/disc and compared with standard antibiotic cephadrine. It was found that at the concentration of 30 μg/disc both the extracts were ineffective against the tested pathogens. Whereas, the same extracts showed moderate to good activity at concentration of 200 μg/disc. The leaf extract exhibited maximum zone of inhibition against *Shigella dysenteriae*.

Ranjith Vimalraj *et al.*, 2009 observed both aqueous and alcoholic extracts of *Cassia fistula* exhibited antibacterial activity against *Staphylococcus aureus*. However, the activity was significantly lower than that of chloramphenicol. There was no inhibition of *B. anthracis* and *B. subtilis*. None of the Gram negative bacteria tested viz., *E.coli, Pasteurella multocida, Salmonella typhimurium* were inhibited by the extracts.

Minimum inhibitory concentrations of different extracts of *Cassia fistula* were tested against fungi and the results revealed that ethyl acetate extract alone showed highest activity against six fungal organisms. The methanol extract from the leaves of *Cassia fistula* had 100% antifungal activity at 10 mg/ml against *Trichophyton rubrum, Microsporum gypseum* and *Penicillium marneffei* (Phongpaichit *et al.*, 2004).

Crude methanol extracts from leaves of *Cassia alata, Cassia fistula* and *Cassia tora* were investigated for their antifungal activities on three pathogenic fungi (*Microsporum gypseum, Trichophyton rubrum* and *Penicillium marneffei*). Among 3 species, C. alata was the most effective leaf extract against *T. rubrum* and *M. gypseum* (Phongpaichit *et al.*, 2004).
Cassia alata leaf powder was used to obtain five extracts which contain anthraquinone compounds in different forms i.e. anthraquinone aglycone extract, anthraquinone glycoside extract, anthraquinone aglycones from glycosidic fraction, crude ethanol extract, and anthraquinone aglycone from crude ethanol extract. All extracts were tested against clinical strain of dermatophytes: Trichophyton rubrum, T. mentagrophytes, Epidermophyton floccosum, and Microsporum gypseum by diffusion and broth dilution techniques to find out the active form for antifungal activity. Thin layer chromatography was developed to demonstrate the fingerprints of chemical constituents of each extract. This investigation pointed out the best in vitro antifungal activity of anthraquinone aglycones from glycosidic fraction qualitatively and quantitatively, compared to other extracts (Mansuang Wuthiudomlert et al., 2010).

Free radicals have significant effects on the structure and general function of the cell. They play a role in the body’s defense against infection by microorganisms. At the same time, they cause a number of diseases by inducing damage to DNA and other important biomolecules. Hence, the search for additional free radical scavengers or antioxidants, especially from plant sources, is of prime importance. Leaf extracts from Cassia alata L., traditionally used for the treatment of a variety of diseases, were evaluated for free radical scavengers using the diphenyl picryl hydrazyl hydrochloride (DPPH) assay (Rajagopal et al., 2014).
In our study DPPH activity of the methanolic extract of *C.roxburghii* flower at 150 µg of concentration showed highest inhibition and minimum inhibition was observed at 10 µg concentration. Inhibitory activity increased with the increasing concentration of the methanolic extract of *C.roxburghii* flowers.

In the present study showed the *C.roxburghii* and *C.auriculata* extracts showed at the highest inhibition than the standard Glucobay (STD). *C.occidentalis* showed moderate inhibition. *C.alata* and *C.fistula* showed minimum inhibition when compared to that of standard. *C.roxburghii* and *C.auriculata* extracts showed the maximum inhibition than the standard. *C.alata* and *C.fistula* showed least inhibition and *C.occidentalis* showed moderate inhibition. *C.roxburghii* and *C.auriculata* extracts showed the highest inhibition than the standard. *C.alata* and *C.fistula* showed minimum inhibition and *C.occidentalis* showed moderate inhibition when compared to that of standard.

*C.auriculata* and *C.roxburghii* extracts showed the highest inhibition than the standard BHT. *C.alata* and *C.fistula* showed moderate inhibition and *C.occidentalis* showed minimum inhibition when compared to that of standard. *C.auriculata* and *C.roxburghii* extracts showed the highest inhibition than BHT. *C.alata* and *C.fistula* showed moderate inhibition and *C.occidentalis* showed minimum inhibition when compared with standard. *C.auriculata* and *C.roxburghii* extracts showed the highest inhibition. *C.fistula* and *C.alata* showed moderate inhibition and *C.occidentalis* showed minimum inhibition.
When Thambidurai et al., (2010) compared the antioxidant activity of flower, fruit and ascorbic acid by DPPH method he showed that the flower exhibited best significant value than standard ascorbic acid.

Michael et al, (1977) reported the presence of aliphatic acid esters, terpene and diterpene alcohol in the leaves of C.auriculata. Diterpene alcohol was the major chemical group in of C.auriculata fractions. 3-0-Methyl-Dglucose (3-0MG), a nontoxic nonmetabolizable derivative of glucose, is effective in reducing the toxicity of streptozotocin (SZ). It has been found to possess antitumor, oncogenic, and diabetogenic properties [Michael et al., 1977]. In the last decades, α-tocopherol has been consecrated as being one of the most efficient antioxidant and radical scavenger.

In previous result they concluded from the data that CFEt significantly reduces the levels of serum and tissue lipids, which are actively raised in streptozotocin diabetes rats. CFEt has beneficial effect on plasma insulin and hexokinase activity. Moreover its antihyperlipidaemic effect could represent a protective mechanism against the development of atherosclerosis (Pari and Latha, 2002a)

In the present study of Lipid Peroxidation of methanolic extract of C.roxburghii flower was tested. α-tocopherol was used as a standard. C.roxburghii flower extract showed maximum inhibition at 150 µg concentration when compared with standard. The activity was minimum at lower concentration. The lipid peroxidation activity increased with the increasing concentration of the methanolic extract of C.roxburghii flowers.
Chapter-11.Discussion

The previous result showed the potential of *Cassia fistula* extract to inhibit lipid peroxidation in rat liver homogenate, induced by the FeCl₂–H₂O₂ system. Decrease in lipid peroxidation by *Cassia fistula* may be a result of it scavenging OH produced by FeCl₂–H₂O₂ and H₂O₂ in the reaction system; this is also confirmed by DPPH and NBT scavenging activity. GSH is mainly involved in the synthesis of important macromolecules and in the protection against reactive oxygen compounds GSH was observed in diabetic rats. The decreased GSH content contributes to the pathogenesis of complications associated with chronic diabetic state. The study showed an elevation in GSH level in tissue after extract treatment (Narendra Silawat *et al.*, 2009).

In our result the cytotoxic activity of methanolic extract of *C.roxburghii* flower shows the % of toxicity and viability at different concentration (10 µg, 25µg, 50µg, 100µg &150µg). At lower concentration showed minimum toxicity and maximum percentage of cell viability. As the concentration was increased the toxicity also increased and the percentage viable cells was reduced nearly to 50%.

The MCF-7cel model has been examined extensively to determine the mechanism(s) of estrogen-stimulated growth. The earlier work identified a secreted glycoprotein from MCF-7 cells that was believed originally to be a growth accelerator (Westley and Rochefort, 1980 and Vignon *et al.*, 1986). However, identification of the protein with monoclonal antibodies (Garcia *et al.*, 1985) and the cloning and sequencing of the gene show the protein to be
the enzyme cathepsin D (Augereau et al., 1988). The clinical community has extensively studied the protein as a potential marker of prognosis in node-positive and node-negative breast cancer. In contrast, a protein of unknown function, pS2, was identified in MCF-7 cells by Cham bon's group (Masiakowski et al., 1982, Brown et al., 1984 and Chambon et al., 1984). The protein has, like the PgR, been linked to good prognosis in breast cancer. The estrogen response element for pS2 is used routinely as an analytical system for reporter genes to study the molecular biology of ER activation.

Gupta et al. (2000) studied the effects of methanolic extract of Cassia fistula seed on the growth of Ehrlich ascites carcinoma and on the life span of tumor bearing mice were studied. They showed an increase of life span, and a decrease in the tumor volume and viable tumor cell count tumor hosts. Cytological studies have revealed a reduction in the mitotic activity, and the appearance of membrane blebbing and intracytoplasmic vacuoles in the treated tumor cells. Improvement in the hematological parameters following ME treatment, like hemoglobin content, red blood cell count and bone marrow cell count of the tumor bearing mice have also been observed.

Duraipandiyanan et al. (2012) isolated rhein from C. fistula flowers and tested against human colon adenocarcinoma cell line COLO 320 DM and normal cell line VERO. Rhein exhibited minimal cytotoxic effect toward VERO cells and inhibited the cell division of carcinoma cells. The compound rhein was identified by spectroscopical method.
The alcoholic extract of *C. occidentalis* inhibited the multiplication of cancer cell lines (Madhulika and Saxena, 2010).

GC-MS method is a direct and fast analytical approach for identification of terpenoids and steroids and only few grams of plant material is required. The importance of the study is due to the biological activity of some of these compounds. The present study, which reveals the presence of components in *Cassia roxburghii* suggest that the contribution of these compounds on the pharmacological activity should be evaluated. The more precise information in qualitative analysis can be obtained by gas-chromatography coupled with mass spectrometry (GC-MS) (Cong et al., 2007). For quantitative determination, gaschromatography with flame ionization detector (GC-FID) and GC-MS are preferred (Lee et al., 2005; Lampronti et al., 2006; HaznagyRadnal et al., 2007).

A total of 7 phytochemicals were identified. The first phytochemical had a retention time of 15.3 seconds and the last compound had a retention time of 26.25 seconds. The second compound 1,1'-dodecylidenebis 4-methyl-cyclohexane had a retention time of 17.2 seconds and the peak area was 13.45%.

Phytol is one among the seven compounds of the present study. Similarly Maria Jancy Rani *et al.* (2011) observed the presence of phytol in the leaves of *Lantana camara* and Sridharan *et al.* (2011) in *Mimosa pudica* leaves. Similar result was also observed in the leaves of *Lantana camara* (Sathish kumar and Manimegalai, 2008). Phytol was observed to have antibacterial
activities against *Staphylococcus aureus* by causing damage to cell membranes as a result there is a leakage of potassium ions from bacterial cells (Inoue *et al.*, 2005). Phytol, Phenol, 2, 4-bis (1-phenylethyl) - which are all have medicinal properties. Phytol is a key acyclic diterpene alcohol that is a precursor for vitamins E and K1. It is used along with simple sugar or corn syrup as a hardener in candies. Mangunwidjaja *et al.* (2006) reported the main components of 9, 12 octadecadienoic acid, Octadec- 9enoic acid and 9,12-actadecadienoic acid present in *Croton tiglium* seed. These compounds were found to have potential antioxidant and anticancer activities. Hexadenoic acid has earlier been reported as a component in alcohol extract of the leaves of *Kigelia pinnata* (Grace *et al.*, 2002) and *Melissa officinalis* (Sharafzadeh *et al.*, 2011). Parasuraman *et al.* (2009) identified 17 compounds with n-Hexadecanoic acid and Octadecanoic acid as the major compounds in the leaves of *Cleistanthus collinus*. GC-MS analysis of ethyl acetate extract of *Goniothalamus umbrosus* revealed the presence of n-Hexadecanoic acid (Siddig Ibrahim *et al.*, 2009). n-hexadecanoic acid, Hexadecanoic acid, Phytol, 9, 12 - Octadecadienoic acid, 9, 12, 15-Octadecatrienoic acid and Squalene were identified in the ethanol leaf extract of *Aloe vera* (Arunkumar and Muthuselvam, 2009) and *Vitex negundo* (Praveen kumar *et al.*, 2010). Squalene is used in cosmetics as a natural moisturizer. Devi *et al.* (2009) reported that *Euphorbia longan* leaves mainly contained n-hexadecanoic acid and 9, 12- Octadecadienoic acid. These reports are in accordance with the result of this study.
The GC-MS analysis showed the presence of various effective chemical moieties. They reveal excellent antibacterial and antioxidant potential. All the results showed that the plant may be used as a good therapeutic agent.

A number of modern drugs has been isolated from the plants, as plants are the natural pool of therapeutic drug free from the side effects caused by any other non-herbal product. In ancient time, almost all disease treatment are managed by plant products. Cassia is an important and potential medicinal plant. The offered literature is about the substantial evidences on the antibacterial activities of its pod and seed extracts. It has been reported that Cassia possess antioxidant, antmutagenic, antitumor, hepatoprotective, antitussive, antimicrobial, anti-inflammatory and so many activities. Its antioxidant activity plays a role in wellness, health maintenance, and the prevention of chronic and degenerative diseases. It is recognized as a rich source of vitamins, flavanoids, tannins glycerides, phospholipids, carbohydrates. Hence, the extract can be utilized for pharmaceutical formulations. The present work summarizes importance of this plant and their action which can be helpful for further investigation to achieve lead molecules in the search of novel herbal drugs. The evaluation of toxic properties is also crucial when considering public health protection because exposure to plant extracts can result in undesirable effects on consumers.
Scientific standardisation and formulation, stability and toxicity testing of the herbal drug compounds, cultivation and conservation of medicinal plants are the present challenges to overcome. Innovative research design protocols and uniform standard guidelines and practices show immense possibilities for the promising future of herbal medicines in health care sector.