The literature pertaining to the study on the “Isolation and Identification of Bioactive Constituent from *Crateva magna* Lour.(DC). and Its Effect on Urolithiasis ” as relevant to the present investigation was reviewed and presented in this chapter under the following headings:

2.1 Pharmacological studies
2.2 Phytochemical studies
2.3 Antioxidant studies
2.4 Antiurolithiatic studies
2.5 Spectral studies

2.1 Pharmacological studies

Tapankumarmaity *et al.* (2000) reported the pharmacognostic profiles of *Cassia tora* L. leaves. The organoleptic characters of fresh leaves showed green colour, whereas the dried leaves showed dark green colour. The odour was disagreeable and the taste was bitter. The fluorescence analysis revealed the presence of different colour variations under UV and visible light. The pharmacological studies on the stem and leaves of *Oxalis corniculata* L. showed acidic odour, light green colour and sour taste. Behaviour of the powder on treatment with different chemical reagents showed different colours under UV and visible light (Mary *et al.*, 2001).

The pharmacognostic constants and phytochemical analysis for the leaf of *Coldenia procumbens* L. were evaluated by Senthamari *et al.* (2002). The organoleptic study of *C. procumbens* leaf exhibited pale greenish grey colour, having characteristic odour and slightly bitter taste. Badami *et al.* (2003) analysed the micro and macroscopical characters of the heartwood of *Caesalpinia sapan*. Heart wood showed orange red colour, the taste was bitter slight odour. The sample powders when treated with various chemicals exhibited various colours in the short, UV and visible light. Jawahar *et al.* (2004) screened the...
pharmacognostical characteristics on *Dodonaea viscosa* L. leaves. The macroscopical characters indicated that the leaf was pale green in colour with characteristic odour and bitter taste. The sample when treated with different chemical reagents, various colours under UV and visible light were observed.

Mazumder *et al.* (2005) have reported the organoleptic and fluorescence analysis of the leaves of *Cassia tora* L. The colour was dark green, the taste was slightly bitter and no characteristic odour was noticed in the leaf. The leaf powders when treated with different chemicals exhibited various colours in the UV and visible light.

Arcus *et al.* (2006) reported that the pharmacognostic characters and phytochemical values of plants could be used as the diagnostic tool for the standardization of *Berteroa incana* (L.) DC. The aqueous extract of *Ficus sycomorus* had inhibitory effect on both smooth and skeletal muscle contractions and contained important constituents for pharmacological activities (Sandabe *et al.*, 2006). The fluorescence analysis was observed in Algerian medicinal plants both under UV and visible light, which showed various colours under ordinary and UV light. Investigations on the dried powders of these plants emphasized that our current knowledge of pharmacognostical actions and preclinical studies updates our knowledge to use them as conventional drugs (Videl *et al.*, 2006).

The pharmacognosy of the leaf of *Mitracarpus scaber* Zucc leaves were studied by Abere *et al.*, 2007. The organoleptic characters of the fresh leaves showed green colour, and was odourless with a slightly acrid taste. The pharmacognostic character of *Hybanthus enneaspermus* studied by Retnam and De Britto (2007) could be used as the diagnostic tool for the standardization of this medicinal plant.

Bhatia *et al.* (2008) screened the pharmacognostical aspects of the seed of *Centratherum anthelminticum* Kuntze. Macroscopic analysis of the seed powder showed dark brown in colour having a characteristic odour and intensely bitter taste. Madhavan *et al.* (2008) conducted the fluorescence analysis along with diagnostic characters on two sources of plants, namely *Convolvulus microphyllus* and *Evolvulus alsinoides*. In fluorescence analysis the powders of both samples treated with different chemical reagents, exhibited different colours in UV and visible light.
Usha kumarai *et al.* (2009) reported that the whole plant of *Acrotrema arnottianum* Wight was tested for powder microscopy, the behaviour of the powdered drug with different chemical reagents and its fluorescence analysis. The whole plant powders had grey olivaceous colour with no characteristic taste and odour and was slightly irritant. The powder was coarse in texture and free flowing.

Various investigations like organoleptic or morphological characters, microscopic or anatomical studies, physico-chemical evaluations (loss on drying, ash values, extractive values) and phytochemical screening were recorded in *Zanthoxylum nitidum* bark. The organoleptic study revealed greenish yellow colour, aromatic odour and bitter taste. The fluorescence analysis showed colour variations under UV and visible light (Bhattacharya and Zaman, 2009).

Sujan Ganapathy *et al.* (2009) reported the macro and micro morphological characters of leaf, quantitative leaf microscopy, fluorescence study of the powder, physico chemical studies and preliminary phytochemical aspects of *Holarrhena antidysenterica* Wall. The organoleptic study revealed pale green colour in leaf and in cream colour bark, whereas the inflorescence showed greenish yellow colour. The powders of the plant samples treated with different chemicals revealed the presence of different colours.

Sindhu *et al.* (2010) studied the microscopic and morphological characters, total ash value, fluorescence analysis and phytochemical screening of *Verbesina encelioides* Benth. root. The fresh roots were light yellowish in colour which turned light brown on drying. The powder showed various colours under UV and visible light, when treated with different chemical reagents. Mahmud *et al.* (2010) investigated the pharmacognostic characters on fresh mature leaves of *Holoptelea integrifolia* (Roxb.) Plach. The organoleptic features of the plant were examined and the result was noted as green colour, slightly aromatic odour and herbal taste. Shruthi *et al.* (2010) carried out the fluorescence analysis of the leaves of *Kirganelia reticulata* Bill. The fluorescence characters of the leaf powder were observed under UV and daylight with various chemicals. It showed fluorescent colours under UV light and showed normal colour under daylight.

Bhuvaneswari *et al.* (2011) conducted the pharmacognostical studies such as extractive values and fluorescence analysis of the leaves of *Thevetia neriifolia* Juss.
Fluorescence analysis of the leaf extracts with different solvents and acids, indicated different colour changes under visible and UV light. Rajan et al. (2011) studied the organoleptic evaluation like taste, odour and colour of the root powder of *Albiziza odoratissima* (L.F) Benth. The result of this evaluation indicated a slightly aromatic odour, dark green colour and characteristically astringent taste.

The organoleptic study of bark and leaf of *Terminalia travancorensis* Wight & Arn exhibited grey colour in bark, green colour in leaf, whereas the odour was pleasant in both the samples. The samples when treated with different chemicals exhibited various colours in UV light (Lakshmi et al., 2012). Kadam et al. (2012) reported the pharmacognostic parameters of the bark of *Mimusops elengi* L., which mainly consisted of macro morphology and microscopical characters, physio-chemical constants and phytochemical screening. The results revealed that the bark was brown in colour, non aromatic and astringent. The fluorescence analysis showed that the powder had various colour variations under UV and visible light. Shanthi et al. (2012) evaluated the organoleptic characters of *Morinda tinctoria* Roxb. Powdered roots were brown in colour, sweet in taste and odour was strong, distinct and pleasant.

Sharma and Pracheta (2013) studied the fluorescence characters of the leaves of *Euphorbia neriifolia*. The fluorescence characters were observed under short and long UV and visible light with various chemicals. The result showed different colour changes under UV and visible light. Lalitharani et al. (2013) performed microscopic, physicochemical constants such as ash, extractive values and fluorescence analysis on *Zanthoxylum rhesta* stem spine. The spine powder showed the characteristic fluorescent green colour when treated with 1N aqueous NaOH under short UV light.

Singh et al. (2014) observed the macroscopy, microscopy, physicochemical evaluation, powder analysis and phytochemical screening of whole plant of *Achyranthes aspera*. The powder showed characteristic behaviour with different chemical reagents under UV and visible light. Sundarraju et al. (2014) reported that *Ocimum basilicum* L. had significant effects on multiple biological systems. Macroscopic study revealed woody green stems, flowers were violet in simple or much branched racemes, odour was aromatic, smell was pleasant, taste was sweet. Behaviour of the powder was determined under UV
and visible light with different chemical reagents, which showed different colour variations. It helped to differentiate from the closely related species of *Ocimum basilicum* L. Partha and Rahaman (2015) screened the pharmacognostic studies of *Adenanthera pavonina* L. In fluorescence analysis powder treated with different chemical reagents gave characteristic colour, when seen under UV light and it was compared with the colour observed under ordinary light.

### 2.2. Phytochemical studies

Phytochemical and antimicrobial activities of leaf and stem extract of *Gynandropsis gynandra* and *Buchholzia coriacea* were analysed. The main secondary metabolites indicated in both the plants were alkaloids, glycosides and steroids. Anthraquinones were slightly indicated (Ajaiyeoba, 2000). Preliminary phytochemical study on different extracts of the roots of *Tragia involucrata* L. was performed. Results showed the presence of alkaloids, reducing sugars, tannins, proteins, flavonoids, sterols, saponins and anthraquinones (Dash *et al.*, 2000).

Gopalakrishnan and Venkataraman, (2000) evaluated the phytochemicals of the root of *TriantHEMA decandra* L. The extracts revealed the presence of various phytochemicals such as steroids, triterpenoids, reducing sugars, tannin, alkaloids, phenolic compounds, flavonoids, anthraquinones and aromatic acids. Verma *et al.* (2000) identified the important phytochemical constituents in the leaves of *VITex negundo*. The results showed the presence of alkaloids, sterol and tannin in the petroleum ether extract. Chloroform extract depicted the presence of flavonoids. Ahmed and Beg (2001) screened the antimicrobial and phytochemical analysis of 45 Indian medicinal plants against multi drug resistance human pathogens. The common phytochemicals present in the plant extracts were have phenols, tannins and flavonoids as major active constituents.

The studies carried out by Senthamari *et al.* (2002) on the preliminary phytochemical tests on various extracts of the leaf of *Coldenia procumbens* L. showed the presence of glycosides, phytosterols, proteins, amino acids, fixed oils, flavonoids, gums and mucilage. Parimala *et al.* (2003) conducted the preliminary phytochemical screening on the methanol extract of *Cleome viscosa*. The result revealed the presence of active constituents such as steroids, saponin, alkaloid, tannin and reducing sugar. The preliminary
phytochemical analysis of leaves of *Atlantia monophylla* indicated the presence of carbohydrates, glucosides, lipids, flavonoids and alkaloids (Manimaran *et al.*, 2003). Morozowska and Wesołowska (2004) screened the preliminary phytochemicals of the plant *Primula veris* L. from field cultivation and also *in vitro* culture. Phytochemical analysis (2D-TLC) revealed that the flavonoid compounds in leafy shoots from *in vitro* culture were similar to those in leaves from field cultivation. The medicinal plants namely *Cleome rutidosperma, Emilia coccinea, Euphorbia heterophylla, Physalis angulata, Richardia bransitensis, Scoparia dulcis, Sida acuta, Spigelia anthelminia, Stachytarpheta cayennensis* and *Tridax procumbens* were tested for phytochemicals. All the plants were found to contain alkaloids, tannins and flavonoids except the absence of tannins in *S. acuta* and flavonoids in *S. cayennensis* (Edeoga *et al.*, 2005).

Skocibusic *et al.* (2006) in their study recorded phytochemical composition and antimicrobial activities of the essential oils from aerial parts of *Satureja subspicata* Vis. Tadhani and Subhash (2006) have confirmed the presence of some active chemical constituents like tannins, alkaloids, saponins, sterols as well as palmitic acid and linolenic acid in *Stevia rebaudiana* leaf samples. Okwu and Josiah (2006) revealed the presence of alkaloids, saponins, tannins in two Nigerian medicinal plants namely *Aspillia africana* and *Bryophyllum pinnatum*. Joseph and Nibeditaya (2006) studied the preliminary phytochemical analysis of leaf powders of *Andrographis paniculata, Aloe vera, Parthenium hysterophorus*. The study revealed the presence of active constituents like cellulose, protein, fat and oil, flavonoids, saponin, sugar, steroid, phenol, quinine and tannin.

The extracts of *Plectranthus glandulosus* were screened by Egwaikhide and Gimba (2007) by using different solvents for secondary metabolites. The extracts revealed the presence of alkaloids, tannins and flavonoids. The aqueous and ethanolic extracts of *Olaus subscorpioidea* stem revealed the presence of alkaloids, steroids and flavonoids (Ayandele and Adebiyi, 2007). Mallikharjuna *et al.* (2007) revealed the presence of alkaloids, flavonoids, glycosides, lignins, phenols, saponins, sterols and tannins in *Strychnos potatorum* root, stem bark and seeds. Doughari *et al.* (2008) confirmed the presence of saponins, tannins, alkaloids and flavonoids in *Senna obtusifolia* (L). They have also concluded that *S. obtusifolia* (L.) can be used to source antibiotic substances for possible
treatment of bacterial and fungal infections including gonorrhea, pneumonia, urinary tract and some mycotic infections. The aqueous and methanol extracts along with dry powders of leaf and bark of the plant *Aquilaria agallocha* Roxb. indicated the presence of alkaloids, anthraquinones, triterpenoids, tannins, fixed oils, fats and glycosides in methanol extracts and saponins, fixed oils, fats, alkaloids and triterpenoids were also found in the aqueous extracts (Dash *et al*., 2008).

Obianime and Uche (2009) reported the presence of flavonoids, tannins, alkaloids, terpenoids, steroids, saponins and cardiac glucosides in the leaves of *Phyllanthus amarus*. Phytochemical screening of ethyl acetate and methanol extracts of *Syzygium cumini* seed revealed that the crude extracts contained alkaloids, flavonoids, glycosides, phytosterols, saponins, steroids and terpenoids. *S. cumini* seeds also have various medicinal values such as anti-inflammatory, anti-diabetic and analgesic activities and also for central nervous system activity (Kumar *et al*., 2009).

The phytochemical profile of *Tridax procumbens* L. revealed the presence of alkaloids, carotenoids, flavonoids, saponins and tannins and a likelihood of the plant serving as a potential supplement and pro vitamin A to the population (Ikewuchi Jude *et al*., 2009). Imaga *et al*. (2010) in a study to screen the aqueous methanol leaf extracts of *Parquentina nigrescens* and *Carica papaya* confirmed the presence of folic acid, vitamin B12, alkaloids, saponins, glycosides, tannins and anthraquinones. Lachumye *et al*.(2010) reported that the methanol flower extract of *Etlingera elatior* contained flavonoids, terpenoids, saponins, tannins, carbohydrates, alkaloids and anthraquinones.

According to Gaire *et al*. (2011) alkaloids, carbohydrates, saponins, glycosides, phytosterols, resins, phenols, tannins, diterpenes, flavonoids, proteins, and amino acids were present in methanol extracts of *F. auriculata* stem bark. Krishnamurthy and Asha (2011) have reported the presence of carbohydrates, proteins, tannins, saponins, terpenoids, flavonoids, steroids, glycosides, alkaloids, phenols and lignin in hot and cold extracts of *Memecylon umbellatum* leaves. Sharma *et al*. (2012) studied the methanolic extract of root, stem and leaf of *Jatropha curcas* L. and revealed the presence of alkaloids, saponins, tannins, terpenoids, steroids, glycosides, phenols and flavonoids. The preliminary phytochemical screening of *Boerhavia diffusa* showed the presence of alkaloids,
flavonoids, steroids, terpenoids, reducing sugars, saponins, tannins, cardiac glycosides and anthraquinones in varying amounts (Apu et al., 2012).

The qualitative analytical data of Citrullus colocynthis seed extracts documented the presence of flavonoids (Benariba, 2013). Preliminary phytochemicals in the leaf of Adhatoda vasica indicated the presence of phenols, tannins, alkaloids, anthraquinones, saponins, flavonoids, aminoacids and reducing sugars (Karthikeyan et al., 2013). Phytochemical and antimicrobial activity of the aqueous and methanol extract of Zingiber officinale, Curcuma longa, Commiphora molmol and Pimpinella anisum, revealed the presence of carbohydrates and saponins in all the samples. Alkaloids were found in Zingiber officinale, where as flavonoids in Curcuma longa and Pimpinella anisum. Steroids and tannins were found only in Zingiber officinale and Curcuma longa respectively (Al-Daihan et al., 2013). Preliminary phytochemical screening of aerial parts of Nelumbo nucifera flowers showed positive results for the presence of flavonoids, alkaloids, phenols, glycosides, carbohydrates and tannins (Krithika et al., 2013).

Butanol and hexane extracts of Parkinsonia aculeata L. leaves were found to possess potent antioxidant and free radical scavenging activity (Sharma and Vig, 2014) which might be due to the presence of flavonoids and polyphenols. Yadav et al.(2014) screened the phytochemicals of the selected six medicinal plants namely, Ficus religiosa, Citrus limonia, Phoenix dactylifera, S. indicum, Swertia chirata and Raphanus sativus. All the selected medicinal plants were found to contain tannins and flavonoids. Alkaloids were present in all the selected plants except F. religiosa, P. dactylifera and R. sativus. Proteins were present only in F. religiosa and S. chirata. Terpenoids were also present in all the selected plants except P. dactylifera. On the other hand, saponins and steroids were absent in the plants except S. chirata and phlobatannins were absent in all the plants except R. sativus.

Preliminary phytochemical screening was done in different parts (leaf, stem and bark) of Diospyros mespiliformis extracted with different solvents (methanol, ethyl acetate and hexane). The result revealed the presence of tannin, saponin, alkaloids, flavonoid, glycosides in various extracts of various plant parts (Ebbo et al. 2015). Devhade et al.(2015) tested the preliminary phytochemicals of Merremia dissecta leaf extracts.
The phytochemical screening showed the presence of alkaloids, glycosides, phytosterols, saponins, tannins and steroids. Amin et al. (2015) detected the phytochemicals of the methanol extract of *Ardisia solanacea* plant. From the result it was concluded that the plant possess various phytochemicals such as alkaloids, carbohydrates, saponins, phytosterols, tannins, phenols, flavonoids, proteins and aminoacids.

### 2.3 Antioxidant studies

Scartezzini and Speroni (2000) worked on *Emblica officinalis* L., *Curcuma longa* L., *Mangifera indica* L., *Momordica charantia* L., *Santalum album* L., and *Swertia chirata* Buch-Ham and reported that these plants are rich in antioxidants. Toit et al. (2001) investigated most of the health benefits of balck, green and oolong teas made from *Camellia sinensis* and attributed to their antioxidant content. One or two cups of tea would provide a similar amount of radical scavenging capacity as five portions of fruits and vegetables or 400 mg vitamin C equivalents. The antioxidant properties of 90% ethanol extracts of leaves and 90% methanol extracts of stem bark, pulp and flowers from Indian laburnum (*Cassia fistula* L.) were investigated. The stem bark had more antioxidant activity in terms of reducing power, inhibition of peroxidation and DPPH radical scavenging ability when compared to pulp and flowers (Siddhuraju et al., 2002). Auddy et al. (2003) observed the antioxidant activity of three Indian medicinal plants, traditionally used for the management of neurogenarative diseases. The result from the ABTS assay showed that the ethanolic extract of *Sida cordifolia* bark was most potent, followed by *Evolvulus alsinoides* and *Cyanodon dactylon*.

Velazquez et al. (2003) studied the antioxidant properties of six medicinal herbs used in traditional Paraguayan medicine. The methanol extracts from *Aristolochia giberti*, *Cocropia pachystachya*, *Eugenia uniflora*, *Piper species*, *Schinus weinmannifolius* and *Schinus terebinthifolia* protected against enzymatic and non-enzymatic lipid peroxidation in microsomal membranes of rat. *C.pachystachya*, *E.uniflora*, *S. weinmannifolius* and *S. terebinthifolia* showed the highest scavenging activity on the superoxide and DPPH radicals. Rani et al. (2004) revealed that orange, tomato and grapes possess predominant quantities of antioxidants namely SOD, catalase, glutathione peroxidase, reduced glutathione, vitamin C and vitamin A.
The antioxidant activity of lemon balm, oregano and pepper mint were analysed immediately after harvest and after drying. The strongest inhibition of linoleic acid (LA) peroxidation was found for fresh and dried oregano compared to peppermint and lemon balm. The antioxidant activities of vegetables and fruits were varied largely but anthocyanin rich samples exhibited the most potent concentration dependent antioxidant activities, including red lettuce, red cabbage, black bean, gala apple peel, mulberry and jambolan (Hassimotto et al., 2005).

Aquil et al. (2006) screened the antioxidant and free radical scavenging properties of the methanolic crude extracts of Terminalia chebula, Terminalia bellirica, Ocimum sanctum, Cichorium intybus, Punica granatum, Allium sativum, Delonix regia, Mangifera indica, Camellia sinensis, Piper cubeba, Lawsonia inermis and Trigonella foenum-graecum. They found a fair correlation between antioxidant and free radical scavenging activity and phenolic content among the plants. According to Nascimento et al. (2006) the hydro ethanolic extract of Turnera ulmifolia L. leaf exhibited greater lipid peroxidation, than α-Tocopherol. Pourmorad et al. (2006) showed that the whole plant of Melilotus officinalis, contain highest amount of flavonoid and phenolic compounds and exhibited the greatest antioxidant activity. The high scavenging property of M. officinalis may be due to hydroxyl groups existing in the phenolic compound’s chemical structure that can provide the necessary component as a radical scavenger.

Kumaran and Karunakaran (2007) have evaluated the antioxidant activity of Phyllanthus debilis, Phyllanthus niruri, Phyllanthus virgatus, Phyllanthus maderaspatensis and Phyllanthus amarus. Among the five plants Phyllanthus debilis exhibited the highest activity for the selected antioxidant assays. Akinmoladun et al. (2007) analysed DPPH Scavenging activity in methanolic extract of Ocimum gratissimum leaf in different concentrations and maximum activity (84.6%) was in 250 µg/ml.

Hazra et al. (2008) reported that the methanolic extract of Spondias pinnata bark contains large amount of flavonoids and phenolic compounds, exhibiting high antioxidant and free radical scavenging activities. Sabir and Rocha (2008) reported that maximum
activity of free radical scavenging, inhibition of Reactive oxygen species and lipid peroxidation was by aqueous extract of *Phyllanthus niruri*.

Mandal *et al.* (2009) tested the free radical scavenging activity and phytochemical analysis in the leaf and stem of *Drymaria diandra* Blume. *D. diandra* stem showed moderate class of anti-lipid peroxidation against thiobarbituric acid but the leaves had high anti-lipid peroxidation, which might be helpful in preventing or slowing the progress of various oxidative stress induced diseases. *Ocimum sanctum* plants were treated with paclobutrazol (PBZ) and abscissic acid (ABA) to analyze the changes in the enzymatic and non-enzymatic antioxidant responses (Hakiman and Maziah, 2009). Non enzymatic antioxidants like ascorbic acid decreased in the ABA treated plants but increased in the PBZ treated plants. Although both PBZ and ABA treatments considerably increased the \( \alpha \)-tocopherol content, it was more in the PBZ treated plants. Enzymatic antioxidants like ascorbate peroxidase and superoxide dismutase were also increased by the treatments. Catalase activity was increased by both growth regulators.

The free radical scavenging activity of different extracts of *Flemingia strobilifera* (L.) R. Br. leaf and root were analysed. Methanolic extract of root showed maximum activity in DPPH radical scavenging activity, hydroxyl radical scavenging activity and nitric oxide radical scavenging activity compared to leaf (Madan *et al.*, 2010). Gacche *et al.* (2010) screened the enzymatic and non enzymatic antioxidant potential of *Vitis vinifera* L. The enzymatic antioxidants namely catalase, peroxidase, ascorbate oxidase and total phenol were found to be high in ethanol extract of *V. vinifera* fruit. Findings of Nagavani and Raghava Rao, (2010) showed that aqueous flower extract of *Michelia champaca* had maximum enzymatic (catalase, peroxidase and superoxide dismutase ) and non-enzymatic antioxidants (ascorbic acid, tannins, flavonoids, anthocyanins and reduced glutathione ).

Khatun *et al.* (2011) screened the antioxidant status of different parts of *Coleus forskohlii* including root, stem, leaves and tubers. The enzymatic antioxidants(catalase, polyphenol oxidase ,peroxidase and superoxide dismutase) and non enzymatic antioxidants( total phenols, flavonoids and \( \beta \)- caroteins) ,were significantly higher in tubers. Pal *et al.* (2011) indicated that ethanolic extract of *Morinda citrifolia* root proved to be a better scavenger of free radicals . Dhal *et al.* (2012) analysed the non-enzymatic and
enzymatic antioxidant potential of leaf extracts of *Cassia tora* and *Cassia sophera*. *C. tora* has greater antioxidant activity than *C. sophera*. *C. tora* showed higher concentration of SOD and Vitamin C, while *C. sophera* exhibited more of CAT, GPX, GSH and protein content. The results indicated that *C. tora* and *C. sophera* leaves possess antioxidant activities and therefore have therapeutic potential to be used as herbal source of antioxidants. The aqueous extract of *Tinospora cordifolia* bark showed excellent *in vitro* antioxidants due to the presence of high phenolics and ascorbic acid contents (Thakur et al., 2012).

According to Sivakrishnan and Kottaimuthu (2013), the ethanolic extract of aerial parts of *Albizia procera* treated rats showed significant increase in the levels of antioxidant enzyme such as superoxide dismutase (SOD), catalase (CAT) and non enzymatic antioxidant, glutathione (GSH) compared to paracetamol induced rats. *Albizia procera* has significant *in-vivo* antioxidant activity and can be used to protect tissue from oxidative stress. The acetone and methanol leaf extracts of *Acalypha alnifolia* had commendable antioxidant activity namely DPPH, superoxide and ABTS radical scavenging activity than other extracts (Revathy et al., 2013).

DPPH radical scavenging activity and non enzymatic antioxidant activity of methanolic extracts of different parts (leaves, stem and fruit) of *Solanum surattense* were estimated by Yadav et al. (2014). The highest total phenol, total flavonoid, ascorbic acid and radical scavenging activity were recorded in methanolic extract of leaves. From the results it could be stated that *S. surattense* has the potential to serve as the source of alternative natural antioxidants and can be used as a medicine against the diseases caused by free radicals. Sea sotia et al.(2014) evaluated the free radical scavenging and phenolic content of various extracts (methanol, chloroform, ethyl acetate and hexane) of *Trigonella foenum graecum* seeds. Ethyl acetate extract of T. foenum graecum seeds showed highest inhibitory potential. There was a positive correlation between the total phenolic content and the antioxidant activity of extracts. These findings suggest that the fenugreek extracts could act as potent source of antioxidants. Udayaprakash et al.(2015) screened the antioxidant and free radical scavenging activity of methanolic leaf extract of *Cinnamomum iners*. The plant possess maximum amount of Total phenolics and Total flavonoids. DPPH free radical
scavenging activity of methanolic leaf extract recorded the IC$_{50}$ value at the concentration of 15 μg/ml. Among various antioxidant assays performed, maximum inhibition occurred in ABTS assay (99.36%) followed by TBA (95.39%) and FTC.

Rajnikant et al. (2015) reported the DPPH free radical scavenging activity of the different extracts and essential oil of *Murraya koenigii* leaf. The highest free radical scavenging activity was reported in methanol extract followed by ethanol and essential oil. Calaborne et al. (2015) studied the total phenolics and flavonoid content in the leaf and root extracts (ethanol, methanol/ water (70/30) and methanol/ water (50/50) ) of *Armoracia rusticana*. Among the extracts tested, aqueous methanolic (70/30, v/v and 50/50, v/v) extracts of both roots and leaves showed higher total phenol and flavonoid contents and antioxidant capacity. Reenu et al. (2015) analysed the antioxidant potential of sequential extracts of fresh and dried rhizomes of *Curcuma caesia*, using solvents viz., hexane, petroleum ether, benzene, chloroform, ethyl acetate, methanol and water. C. caesia showed significant antioxidant activity in chloroform, benzene and ethyl acetate extracts. The chloroform extract was highly effective as free radical scavengers. The highest total phenol content was also exhibited by chloroform and benzene extracts.

### 2.4 Antiurolithiatic studies

Perez et al. (2000) noticed that the antiurolithiatic activity of two flavonoid compounds isolated from *Eysenhardtia polystachya* namely 7-hydroxyl 2L,4',5L-trimethoxy isoflavone and 7-hydroxy-4L-methoxy isoflavone, was tested in rats by observing calculus formation. A significant decrease in urinary stone size was observed in animals treated with these compounds. Garimella et al. (2001) investigated the effect of aqueous leaf extract of *Melia azadirachta* L. were studied against ethylene glycol induced nephrolithiasis in male wistar rats and they concluded that the treatment with leaf extract reduced urinary calcium, oxalate, phosphate and elevated magnesium levels and urine volume.

Selvam et al. (2001) reported that the increased excretion of calcium, oxalate, uric acid, phosphorus and protein in hyperoxaluric rat is reduced by the administration of *Aerva lanata* and Vedippu chunnam. It increases the urine volume and there by the solubility product with calcium oxalate and other crystallizing salts such as uric acid, which may
induce epitaxial deposition of calcium oxalate. Combination therapy was found to be more effective and this indigenous medicine can be used successfully as an antiurolithiatic agent. Khan et al. (2001) investigated the effect of *Ammi visnaga* seeds on kidney stones and reported that the antilithiatic effect is mainly because of highly potent diuretic activity and amelioration of uraemia and hyperbilirubinemia by seeds of *Ammi visnaga*.

Freitas *et al.* (2002) reported the inhibitory effect of *Phyllanthus niruri* on crystal growth, in a rat model of urolithiasis induced by introduction of calcium oxalate seed in bladder of rats. The effect may be due to higher levels of glycosoamino glycans incorporated into calculi. Al-Ali *et al.* (2003) confirmed the diuretic and contractile effects of *Tribulus terrestris* with its potential of propelling urinary stones. The administration of *Herniaria hirsuta* aqueous extract to experimentally CaOx induced nephrolithiatic rats reduced the deposition of crystals in the kidneys confirming its antilithiatic effects (Atmania *et al.*, 2004). Farroq *et al.* (2004) tested the aqueous seed extract of *Dolichos biflorus* and rhizomes of *Bergenia ligulata* for their *in vitro* antiurolithiatic activity. The extracts were compared with standard cystone. Extracts of *D. biflorus* showed activity almost equivalent to cystone while *B. ligulata* showed less activity.

Edible plant *Triandema monogyna* and the pulse *Macrotyloma uniflorum* extracts were found to be effective in the inhibition of calcium oxalate crystallization (Das *et al.*, 2005). According to Aziz *et al.* (2005), *Plantago major*, potassium citrate and allopurinol do have significant inhibition effects on the size of calcium oxalate crystals. According to Velazquez *et al.* (2005) IC₅₀ values on the size of crystals, *Plantago major* was the best inhibitor on the size of crystals followed by potassium citrate and allopurinol. The aqueous extract of *Zea mays* L. stigma/style (Corn silk) is effective against renal calculi, particularly of calcium oxalate origin. Alvin Jose and Janardhanan (2005) reported that the ethanolic extract of *Plectrathus amboinicus* Lour. has an inhibitory potential on lithiasis. The ethanolic extract, significantly reduced the elevated levels of calculogenic ions in urine and elevated the urinary concentration of magnesium.

Saponin rich fraction isolated from the plant *Herniaria hirsuta* was found to be strongly suppressive in each step of crystal formation, growth and aggregation of CaOx crystals (Fouada *et al.*, 2006). Christina *et al.* (2006) reported that the aqueous and
alcoholic extracts of the root wood of *Melia azedarach* significantly reduced the elevated urinary oxalate, showing a regulatory action on endogenous oxalate synthesis in hyperoxaluria induced with ethylene glycol. According to Karadi *et al.* (2006), the increased deposition of stone forming constituents in the kidneys of calculogenic rats was also significantly lowered by curative and preventive treatment using aqueous and alcoholic extracts. The results indicated that the root-wood of *Moringa oleifera* is endowed with antiurolithiatic activity. Soundararajan *et al.* (2006) have reported that administration of aqueous suspension of *Aerva lanata* reduced oxalate synthesizing enzymes confirming the curative property of it for urolithiasis.

Beghalia *et al.* (2007) reported that the herbal extracts of *Tetraclinis articulata* and *Chamaerops humilis* inhibited the growth of calcium oxalate monohydrate crystals *in vitro*. Touhami *et al.* (2007) found that the administration of lemon juice effectively prevented the development of urolithiasis in rats. These findings support the use of lemon juice as an alternative medicine to prevent urolithiasis.

Beghalia *et al.* (2008) opined that the aqueous extracts of *Ammodaucas leucotrichus* and *Ajuga iva* were found to potently inhibit the nucleation and aggregation phases of calcium oxalate crystallization, while extracts of *Erica multiflora* and *Globularia alypum*, inhibited nucleation and growth of the crystals but not their aggregation. Bahuguna *et al.* (2008) tested the aqueous and alcoholic extracts of *Melia azadirachta* leaves for antiurolithiatic activity against renal calculi administration of ethylene glycol. *M.azadirachta* leaves significantly reduced the elevated urinary stones. The leaf juice of *Sesbania grandiflora* showed significant antiurolithiatic activity against calcium oxalate stones and also exhibited antioxidant properties. The results obtained in this study provide evidence for the efficacy of the leaf juice of *S.grandiflora* as antiurolithiatic agent (Doddola *et al.*, 2008). Laikangbam *et al.* (2009) showed that ethanol extract of *Cassia adnata*, *Cuminum cyminum*, *Hibiscus sabdariffa*, *Oxalis corniculata* and *Piper longum* showed promising roles in prevention and cure of urolithiasis.

The inhibition of mineralization of urinary stone forming minerals by medicinal plants *i.e.* *Achyranthes aspera* L., *Passiflora leschenaultii* DC, *Solena amplexicaulis* (Lam.) Gandhi, *Scoparia dulcis* L. and *Aerva lanata* (L.) have been investigated by Farook
Increased intake of fruit juice and seed extract of the above mentioned plants would be helpful in urinary stone prophylaxis. Bahuguna et al. (2009) indicated that administration of juice and alcohol extracts of *Pyracantha crenulata* fruit to rats with ethylene glycol induced lithiasis reduced and prevented the growth of urinary stones, thus supporting folk information regarding the antiurolithogenic activity of the plant.

Arasaratnam *et al.* (2010) have reported that *Tribulus terrestris* extract has antiurolithiatic activity. Ethanolic extract of *Phyla nodiflora* exhibited significant effect in preventing calcium oxalate stone formation and also in dissolving the pre-formed calcium oxalate stone in the kidney with significant effect on both *in vitro* and *in vivo* (Dodoala *et al.*, 2010). Fahad *et al.* (2010) indicated that the root-wood of *Moringa oleifera* is endowed with antiurolithiatic activity. Hosseinzadeh *et al.* (2010) reported that *Pinus eldarica* aqueous fruit extract prevents calcium oxalate deposition. The protective effect of the hydro-alcoholic extract of roots of *Rubia cordifolia* L. against ethylene glycol induced urolithiasis and its possible underlying mechanisms using male wistar albino rats, indicated that administration of root extract of *R. cordifolia* L reduced and prevented the growth of urinary stones. Therefore it is helpful to prevent the recurrence of this disease as it showed its effect on early stages of stone development (Divakar *et al.*, 2010). The ethanolic extract of whole plant of *Phyla nodiflora* L. for its antiurolithiatic activity against most common type of renal stone (calcium oxalate) was examined by Sujatha *et al.* (2010). The results revealed that the plant possess significant antiurolithiatic activity.

The antiurolithiatic activity of *Coleus aromaticus* leaves in ethylene glycol induced urolithiatic rats was reported by Venkatesh *et al.* (2010). The plant possess antiurolithiatic activity and significantly reduced the stones. Histopathological reports also confirmed that chronic administration (300 & 600 mg/kg) diminished the number of calcium oxalate crystals in kidneys. The aqueous extract of *Hygrophila spinosa* (200 mg/kg) was administered orally against the ethylene glycol induced urolithiasis in rats by Satish *et al.* (2010) significantly reduced the elevated levels of these ions and proteins in urine. The histological findings also showed significant activity after treatment with extract.

Khan and Pradhan (2011) reported that the hydroalcoholic extract of *Ageratum conzoides* possess significant antiurolithiatic activity. Praveen kumar *et al.*
(2011) investigated the effect of ethanolic extract of *Tinospora cordifolia* (Wild.) Miers on calcium oxalate crystallization in urolithiasis. They found that the *T. cordifolia* stem extract remarkably inhibited the calcium oxalate crystal formation. Rad *et al.* (2011) studied the fraction isolated from *Cyanodon dactylon* and tested for antiurolithiatic activity. The study indicated that administration of *C. dactylon* N-butanol fraction and ethyl acetate fraction showed beneficial effects on prevention and elimination of CaOx calculi in the rat kidney.

*Kigelia africana* fruit extracts showed anti-lithogenic properties in both *in vivo* and *in-vitro* antilithiatic models. An overall result from both the lithiatic models concludes that *K. africana* has a potent anti-urolithogenic activity and would help in renal stone dissolution and elimination (Gupta *et al.*, 2011). *Pergularia daemia* whole plant extract (50% alcohol) was investigated for its antiurolithiatic and diuretic activity. *P. daemia* had positive effect on urolithiasis at a concentration of 400mg/kg body weight (Vyas *et al.*, 2011). Pathan *et al.* (2011) suggested that ethanolic extract of *Coleus barbatus* and *Trigonella foenum graecum* roots and their combination have positive effect on urolithiasis. Kore *et al.* (2011) investigated the hydro alcoholic extract of *Lawsonia inermis* L. leaves which showed significant antiurolithiatic activity against calcium oxalate stones.

Sathya and Kokilavani (2012a) indicated that the ethanolic extract of *Saccharum spontaneum* had potent antiurolithiatic activites in ethylene glycol induced male wistar albino rats. These results showed that the antiurolithiatic effect of the plant may be due to its antioxidant and free radical scavenging properties of the secondary metabolites present in the plant. According to Sathya and Kokilavani (2012 b) the ethanolic extract of *Acalypha indica* has potent antiurolithiatic activities in ethylene glycol induced male wistar albino rats. Manjula *et al.* (2012) reported that the aqueous stem extract of *Costus igneus* inhibit the formation of calcium oxalate crystals. This study was focused on finding a new alternative medicine for the treatment of calcium oxalate urinary stone. According to Mohd and Debasish (2012) the antiurolithiatic activity of hydroalcoholic extract of leaves of *Ceropegia bulbosa* significantly protected animal’s kidney from urolithiasis.

Parmar *et al.* (2012) investigated the effect of methanolic stem extract of *Swertia chirata* in experimentally induced urolithiasis in rats. Histopathological analysis also revealed deposition of calcium oxalate crystals and disruption of tubular cells and
juxtglomerular cells. The deposition and disruption were reduced in rats treated with swertia chirata stem extract. According to Khan and Pradhan (2012) the hydro alcoholic leaf extract of Ceropogia bulbosa showed significant antiurolithiatic activity. Nizami et al. (2012) screened the antilithiatic effect of the whole Leea macrophylla Roxb ethanol extract in ethylene glycol induced urolithiasis model of rats. The results of this study demonstrated very promising anti-urolithiatic effect of L.macrophylla extract with preventive and therapeutic treatments.

The inhibition of in vitro calcium oxalate crystal formation of various extracts of Hyptis suavelones (L) was investigated by Kum kum and Ranjana (2012). Thus the inhibitory potency of alcohol extracts of H.Suavelones (L) was comparable to that of cystone. Diana and George (2012) reported that the bioactive compounds in the root extract of A.aspera have controlled the crystal growth. Tayal et al.(2012) screened the antilithiatic and cytoprotective activity in Terminalia chebula and recorded it as a potential candidate for phytotherapy against urolithiasis, as it not only has a potential to inhibit nucleation and the growth of CaOx crystals, but also has a cytoprotective role. Mi et al.(2012) studied the antiurolithiatic activity of Desmodium styracifolium and Pyrrosiae petiolosa. From the result it was found that medium and high dose of D.styracifolium had beneficial effect in preventing calcium oxalate stone formation when compared to Pyrrosiae petiolosa.

Rajeshwari et al. (2013) reported that the aqueous leaf extract of Convolvulus arvensis showed beneficial effect, when compared to flower extract against urolithiasis, by decreasing CaOx excretion and preventing crystal deposition in the kidney tubules. The extract of Bergenia ciliata was significantly more effective in inhibiting the nucleation and aggregation of COM crystals in a dose-dependent manner. Moreover, the extract induced more CaOx dihydrate crystals, with a significant reduction in the number and size of COM crystals (Saha and Verma, 2013). The Ethanolic extract of solanum virginiamum (200 mg/kg, 400 mg/kg) was tested on ethylene glycol induced urolithiasis in rats. Treatment with Ethanolic extract of solanum virginiam significantly reduced the elevated levels of ions in urine as well as blood urea nitrogen, serum creatinine and serum uric acid level (Chinnala et al., 2013). Kishore et al. (2013) concluded that the ethanolic extract of portulaca oleracea is effective against ethylene glycol and ammonium chloride induced urolithiasis in albino rats.
Ahmed et al. (2013) have showed that hydro alcoholic extract of Adiantum capillus veneris L. significantly reduced the number of crystals. Serum levels of calcium, phosphorous and blood urea were also found to be decreased in extract treated group. Vijaya et al. (2013) evaluated the antiurolithic activity of methanol leaf extract of Glochidion velutinum. The levels of BUN, creatinine and uric acid significantly reduced in plant extract treated groups when compared to ethylene glycol treated groups. The results indicated that the dried leaves of G. velutinum have antiurolithic activity. Takawale et al. (2013) reported that Lagenaria siceraria fruit powder showed significant antiurolithic activity. Baheyti and Kadam (2013) reported the antiurolithic activity of a poly herbal formulation against calcium oxalate induced urolithiasis in rats. They concluded that the daily oral treatment with polyherbal formulation significantly decreased, the quantity of calcium oxalate deposited in the kidney.

Galani and Panchal (2014) reported that 70% methanolic extract of Centratherum anthelminticum seeds had the higher capacity to inhibit the crystal formation and aggregation. They suggested possible antinephrolithic activity of C. anthelminticum seeds against calcium oxalate stones. Patel et al. (2014) studied antiurolithic activity of Withania somnifera in ethylene glycol induced urolithiasis in rats. After completion of treatment after 24 hrs urine was collected and blood was collected by retro orbital puncture and kidney histopathology was done. The methanolic extract of Withania somnifera showed increased urinary volume and pH. All the treated groups showed decrease in calcium, oxalate, phosphate, creatinine, urea and uric acid level in EG induced urolithiasis.

Lingam et al. (2014) studied the antiurolithic activity of the aqueous and alcoholic extracts of Melia azadirachta L. leaves in calcium oxalate urolithiasis of male albino rats. Treatment with aqueous or ethanol extract (250mg/kg) significantly reduced the elevated levels of calcium, oxalate and phosphate excretion in urine. Both the extracts revealed that Melia azadirachta L. leaves have potent antiurolithic activity. Sandhyarani et al. (2014) found that the methanolic leaf extract of Cochlosperum vitifolium significantly decreased the calcium, phosphorous, oxalate and magnesium in glycolic acid induced groups. It also increased the urine volume, thereby reducing the tendency for crystallization.
Antiurolithiatic activity of *Centratherum anthelminticum* (L.) Kuntze seeds against ethylene glycol induced urolithiasis in rats was tested by Varsha and Rital (2014). Treatment with aqueous extract of *C. anthelminticum*, significantly reduced stone forming promoters, (calcium, oxalate, phosphate, creatinine, urea, uric acid) and enhanced stone forming inhibitors (magnesium) with significant antioxidant activity in a dose dependent manner. Ramesh *et al.* (2014) analysed the ethanolic leaf extracts of *Orthosiphon stamineus* used for antiurolithiatic activity. The ethylene glycol was used to induce urolithiasis in albino rats. The final results showed that the ethonolic extracts of plants had good activity when compared with the standard drug.

### 2.5 Spectral studies

The flavonoid compounds namely 7-hydroxy-2′,4′,5′-trimethoxyisoflavone and 7-hydroxy-4′-methoxyisoflavone, isolated from aqueous heartwood extract of *Eysenhardtia polystachya* showed antilithiatic and diuretic activity. Chemical analysis of the stones revealed that both compounds reduced calcium and magnesium concentrations. The results support the traditional use of the plant against urolithiasis (Perez *et al.*, 2000). Fouada *et al.* (2006) obtained fraction F15 compound, identified as saponins by preliminary fractionation of *Herniaria hirsuta* and assumed that this could be responsible for the treatment of kidney stone formation.

Saponin rich fraction isolated from the plant *Herniaria hirsuta* was tested for antiurolithiatic activity *in vitro* and *in vivo*. In *in vitro* condition by increasing the doses, (0.2 to 1.0 mg/ml) the size of the CaO\text{X} crystals were reduced when compared to control. In *in vivo* condition the dose 5mg/day, reduced significantly crystal deposition in lithiatic rats. Saponins may be responsible for the beneficial effect of *Herniaria hirsuta* in the treatment of kidney stones (Fouada *et al.*.,2006). GC-MS analysis of *Carica papaya* *L*. leaf showed the presence of 5,7-Dimethoxycoumarin and polar molecules such as protocatechuic acid, p-coumaric acid, caffeic acid, chlorogenic acid, kaempferol and quercetin (Canini *et al*. ,2007).

GC-MS analysis of the dichloromethane extract of the bulbs of *Ornithogalum cuspidatum* Bert. by Nazifi *et al.* (2008) revealed the presence of 25 steroids. Fungisterol was found to be the most abundant steroid in the dichloromethane extract.
Gallo *et al.* (2008) studied the HPTLC analysis of methanol extract of *Lawsonia inermis* L leaf. The major significant spots appeared at about R$_f$ 0.46 to R$_f$ 0.62. The GC-MS analysis of *Silene armeria* L. revealed the presence of 28 compounds, which represented 89.03%, possesses a wide range spectrum of fungicidal activity and could become an alternative to synthetic fungicides for controlling certain important plant fungal diseases (Bajpai *et al.*, 2008). Several phytochemicals which could be responsible for antiurolithiatic effects are flavonoids and terpenoids (Arafat *et al.*, 2008), saponins and tannins (Doddola *et al.*, 2008). The HPTLC analysis of the aqueous acetone extract of *Osbeckia aspera* leaves contain phenolic and terpenoid compounds (Grayer *et al.*, 2008).

Uma *et al.* (2009) revealed the GC-MS analysis of methanol extract of *Cinnamomum zeylanicum*. The most identified compounds by GC-MS to have antimicrobial property were monoterpenes, sesquiterpenes, aromatic aldehydes and ketones. Cinnamaldehyde was the major compound responsible for the antimicrobial activity. Sasikumar *et al.* (2009) reported that the methanolic extract of *Pandanus odoratissimus* L. root was examined for their contents of phenolics using HPTLC method. Blue and brown colour zones were detected in UV after derivatization in HPTLC which confirmed the presence of polyphenols. The R$_f$ value of the root extract was found to be from 0.24 to 0.70.

β-sitosterol-D-glycoside was isolated from the petroleum ether leaf extract of the leaves of *Ocimum sanctum* L and the structure was elucidated with UV, IR, 1H-NMR, 13C-NMR, spectral data (Rahman *et al.*, 2009). Abdelwahab *et al.* (2009) studied the GC/MS analysis of ethyl acetate leaf extract of *Goniothalamus umbrosus* which revealed the existence of 1-butyl-2-cyclohexen-1-ol (46.84%), benzaldehyde (4.42%) and globulol (4.07%). Samejo *et al.* (2009) reported a compound namely Maltol (3-hydroxy-2methyl-4H-pyran-4-one) which was isolated from leaves of *Abies pindrow* by column chromatography and its molecular structure was identified using NMR spectroscopy.

Column chromatographic analysis of ethyl acetate cone extract of *Metasequoia glyptostroboides* resulted in isolation of an abietane-type diterpenoid, taxodone (Bajpai and Kang, 2010). The two compounds namely solasonine and solasodine isolated from the methanolic extract of *Solanum xanthocarpum* berries, showed antiurolithiatic activity.
The isolated solasonine had a greater antiurolithiatic activity compared to solasodine (Patel et al., 2010). Byahatti et al. (2010) reported that leaves of *Bergenia ciliata* possess antiurolithiatic property, as they dissolve experimentally prepared kidney stones, calcium oxalate and calcium phosphate by an *in-vitro* model.

Two flavonoids, a triterpenoid and a mixture of sterols were isolated, namely oleanolic acid, β sitosterol, campesterol, isoquercetin (quercetin-3-O-β-D-glucose) and kaempferol from the bioactive ethyl acetate and n butanol soluble parts of successive methanol extract of fruits of *Lagenaria siceraria*, when subjected to column chromatography. Their structures were elucidated by means of UV, IR, mass, and NMR and the compounds were identified by comparison of their $^1$H-NMR, $^{13}$C-NMR and mass spectra with the literature. These compounds may explain the medicinal value of fruits of *Lagenaria siceraria* (Gangwal et al., 2010). Derwich et al.(2010) screened the leaves of *Mentha pulegium* by GC-MS and identified totally twenty eight constituents. The major component was piperitone (35.56%). Patel et al.(2010) tested the solasonine a glycoalkaloid and solasodine a steroidal compound isolated from the methanolic extracts of *Solanum xanthocarpum* for antiurolithiatic activity and found that solasonine had greater antiurolithiatic activity compared to solasodine.

Chemical analysis of aerial parts of *Ixora paviflora* indicated b-sitosterol, b-sitosterol-b-D-glucoside, kaempferol and kaempferol-7-O-methyl ether. Further the structures of these compounds were determined by IR, UV, NMR spectroscopy (Bachheti et al., 2011).HPTLC finger printing of ethanolic extract of *Naringi crenulata* stem revealed 10 spots with R$_f$ values in the range of 0.07-0.63 and ethanol extract of lead revealed 8 peaks with R$_f$ values in the range of 0.09 to 0.49 (Sampathkumar and Ramakrishnan, 2011). Kamboj and Saluja (2011) reported that the dried aerial parts powder of *Ageratum conyzoides* yielded white crystalline powder which was subjected to physical, chemical and spectral identification by IR, $^1$H-NMR, $^{13}$C-NMR and GC-MS. The compound was confirmed as stigmasterol and β-sitosterol.

Abirami and Rajendran, (2011) reported the compounds namely, tetradecanoic acid (39.70) and 2methoxy -4α- methylanrost-2-en-17-1-one 5β (24.38) from the methanol extract of *Indigofera aspalathoides* that have been evaluated using GC- MS. Yamunadevi
et al. (2011) recorded HPTLC profile of terpenoids of *Aerva lanata* that revealed the presence of twenty seven different types of terpenoids with twenty seven different *R*$_f$ values starting from 0.06 to 0.97. Such finger printing could be useful in differentiating the species from the adulterant and act as a biochemical marker for pharmaceutical industry and plant systematic studies. The ethanolic extract of *Mussaenda frondosa* has been subjected to GC-MS analysis. Twenty chemical constituents have been identified, The major chemical constituents were (-)-Quinic acid (32.87 %), 4-((1E)-3-Hydroxy-1-propenyl)-2-methoxyphenol (8.30%), Naphthalene, decahydro- 2-methoxy-(7.20 %) and 1,2,3-Benzenetriol (7.70%). So it is recommended as a plant of phytopharmaceutical importance (Gopalakrishnan, 2011).

GC-MS analysis of ethanol extract of *Polycarpea corymbosa* (L.) confirmed the presence of thirteen phytocompounds that justifies the use of the whole plant for various ailments by traditional practitioners. (Balamurugan et al., 2012). HPTLC analysis of ethanol extract of *Zanthoxylum rhetsa* (Roxb.) DC fruit showed the presence of eight glycosides, ten flavonoids, six essential oils, five anthraquinones, seven coumarins and eight terpenoids (Alphonso and Saraf, 2012). Sule et al. (2012) reported that the acetone leaf extract of *Olax mannii* Oliv. led to the identification of two compounds namely glutinol and rhoiptelnon using GC-MS and their structures were elucidated on the basis of one- and two-dimensional NMR spectroscopy.

Sharifa et al. (2012) studied the antiurolithic compound from *Plantago major* L. From the results they concluded that the active compound in methanol extract of *Plantago major* inhibited the area of formation, and size of calcium oxalate crystals, was a compound terpenoid and effect of that terpenoid compound was much better than that of zyloric and potassium citrate. Monika et al. (2012) reported that the alkaloid compound cerpegin isolated from the root of *Ceropegia bulbosa* var. Lushii was for antiurolithic activity. The compound showed maximum dissolution of calcium oxalate and calcium phosphate in comparison to extract treated group. Patra et al. (2012) screened the $^1$H NMR analysis and bioautography screening of methanol extract of *Excoecaria agallocha* L. The $^1$H NMR spectroscopic analysis of the plant extract revealed the presence of acyclic aliphatic and $\alpha$-mono substituted aliphatic group of compounds in the sample. Abirami and Rajendran
Selvaraj et al. (2013) reported that two flavonoids namely rutin and quercetin were isolated from the methanol extract of *Azolla microphylla* and identified by NMR. Sushma et al. (2013) screened the HPTLC fingerprinting of leaf extracts of *Ficus nervosa* Heyne ex Roth. HPTLC finger printing of chloroform extract of leaf revealed 11 peaks with *R*[ sub]f values in the range of 0.07 to 1, ethyl acetate extract of leaf showed 11 peaks with *R*[ sub]f values in the range of 0.07 to 0.99 and 90% ethanolic extract of leaf revealed 13 peaks with *R*[ sub]f values in the range of 0.03 to 1. These can be used as a diagnostic tool for the correct identification of the plant and it is useful as a phytochemical marker and also a good estimator of genetic variability in plant populations.

Mariajanyrani et al. (2013) reported that the compound oleananoic acid a terpenoid extracted from the leaves of *Bougainvillea glabra* showed excellent good antibacterial activity. Vyas and Argal (2013) studied the antiurolithiatic activity of ethanolic root extract of *Lantana camera* and also oleanolic acid isolated from roots of *L. camara* in albino wistar male rats .The results indicated that the treatment with oleanolic acid significantly reduced the calcium deposition in the kidney, when compared to plant extract treated group. Atale and Rani (2013) studied the GC-MS analysis of methanolic and ethanolic seed extracts of *Syzygium cumini*. They identified thirty four major compounds in methanolic and 24 major compounds in ethanolic extract. Sasikala (2013) studied the *in silico* antiurolithiatic screening of *Rotula aquatica* Lour. The triterpenoid compound namely 3-O-Acetyl-11-Keto-β-Boswellic acid isolated from the aqueous root extract of *Rotula aquatica* Lour interacted with Tamm-Horsfall protein a least glide score of -2.35. This result indicated the antiurolithiatic property of the compound.

The petroleum ether extract of *Citrus medica* seeds were analysed for the bioactive compounds by GC-MS method. Twenty three bioactive constituents were identified and these constituents are in high concentration of oleic acid with retention time 18.57 (Patil et al., 2014). The presence of some of these bioactive constituents in the plant extract may provide the scientific evidences for the anti fertility activity and contraceptive
properties of the plant. By the use of NMR (\(^1\)H NMR and \(^{13}\)C NMR) and Mass spectroscopy the compound namely piperine, mixture of glycerin and ascorbic acid was identified in 50% ethanolic extract of fruits of \textit{Scindapsus officinalis} (Roxb.) Schott (Velraj et al., 2014).

Zayed et al. (2014) identified compounds in \textit{Neolamarckia cadamba} leaf extracts using GC-MS. A total of 26 compounds were identified and the major chemical constituents were n-hexadecanoic acid (44.88%), hexadecanoic acid ethyl ester (17.96%) and octadecanoic acid ethyl ester (11.71%).

Sivakumar and Divya (2015) studied the bioactive compounds present in the ethyl acetate extract of \textit{Cordia monoica} leaf by GC-MS analysis. The analysis revealed the presence of 20 compounds. Some of the bioactive compounds screened include phytol acetate, n-hexadecanoic acid, neophytadiene, neopentyl hydroxyl acetate and nonacosane. The compounds were identified by comparing with retention time and peak area and by interpretation of mass spectra. The presence of these bioactive compounds in \textit{Cordia monoica} leaves lends credence to its use by the human community. It also holds for the production of novel drugs with isolation of specific compounds.

According to Rajeswari and Rani (2015) the GC-MS analysis of ethanolic root extract of \textit{Lawsonia inermis} showed different peaks with low and high molecular weight determining the presence of 41 phytochemical compounds. The presence of these compounds may proceed to find out various therapeutic activities. The result of the GC-MS analysis of the ethanolic seed extract of \textit{Cola nitida} showed that the isolated caffeine from kolanut contains 82.69 % pure caffeine with 96% in quality (Salahdeen et al., 2015). Dinnimat and Jalalpure (2015) studied the \textit{in silico} antiurolithiatic screening of isolated constituents of \textit{Aerva lanata} (L). Two compounds namely quercetin and betulin isolated from \textit{n}-butanol and hexane fraction of \textit{A. lanata} and their structures were characterized using NMR spectroscopy. These two compounds were studied by \textit{in silico} method against a protein namely 2 ETE of oxalate oxidase and these two compounds have produced significant results which substantiate their claim of bioactivity.

GC-MS analysis of bioactive compounds from the whole plant ethanolic extract of \textit{Evolvulus alsinoides} was tested by Gomathi et al. (2015). GC-MS analysis revealed the
presence of various compounds like piperine, octodecanoic acid, hexadecanoic acid and squalene. These findings support the traditional use of *Evolvulus alsinoides* in various disorders.

Chipiti et al. (2015) screened the GC-MS analysis of the ethanol and aqueous extracts of *Cissus cornifolia* root. The GC-MS analysis of the aqueous and ethanol extracts of the roots indicated the presence of the common aromatic phenolic compounds, pyrogallol, resorcinol and catechol, a fatty acid, n-hexadecanoic acid and an aldehyde, vanillin. According to Singh et al.(2015) the presence of twelve different compounds namely benzene 1, 2, 4, 5-tetramethyl (2.85%), benzene 1, 2, 3, 5-tetramethyl (1.16%), 1-decanol, 2, 2-dimethyl (4.38%), phenol 2, 4-bis (1, 1-dimethyl ethyl) (7.78%), heptadecane (3.60%), 3-hexadecanol (3.30%), i-propyl tetradecanol (3.64%), benzo (h) quinoline (3.66%), n-hexadecanoic acid (6.54%), octadecanoic acid methyl ester (0.81%), phytane (1.95%) and pentadecane (2.25%) were present in the water extract of *Wrightia tinctoria* bark. Alwahsh et al. (2015) isolated the nine flavonoids from aerial parts of *Teucrium barbeyanum*. The compounds were characterized using NMR.