Consumption of fruits and vegetables is shown to lower the risk for chronic diseases such as cancer, cardiovascular diseases and stroke. The positive health effects may be due to high contents of certain phenolic compounds in plant-derived foods. Recently, phytochemicals and their effects on human health have been intensively studied. In particular, a search for antioxidants, hypoglycemic, and anticancer agents in vegetables, fruits, teas, spices and medicinal herbs has attracted great attention.

This chapter encompasses the published literature on medicinal uses and various studies on the genus *Tricalysia* a member of the family Rubiaceae which is chosen for the present study.

### 2.1. Family Rubiaceae

Rubiaceae are a family of flowering plants, variously called the coffee family, madder family or bedstraw family. Members of the coffee family tend to be concentrated in warmer and tropical climates around the world. Currently, about 611 genera and more than 13,000 species are placed in Rubiaceae. This makes it the fourth-largest family of flowering plants by number of species, and fifth largest by number of genera. The group contains many commonly known plants, including the economically important coffee (*Coffea*), quinine (*Cinchona*), and gambier (*Uncaria*), the medicinal ipecacuanha (*Carapichea ipecacuanha*), and the horticulturally valuable madder (*Rubia*), west indian jasmine (*Ixora*), partridgeberry (*Mitchella*), *Morinda*, *Gardenia*, and *Pentas*.

During the survey of Rubiaceous taxa, it was investigated that most of the plants of this family are of great medicinal value. Several ailments like ulcers, dysentery, athlete’s foot, diabetes, whooping cough, bronchitis, asthma, migraine etc. are successfully cured by the use of the plants. Some plants of family Rubiaceae are of miraculous importance which are used in the treatment of snake bite, scorpion sting, regulation of menses and securing the birth of male child. A very poor attention has still been paid on family Rubiaceae regarding its medicinal properties.
The genus *Tricalysia* comprises of about 50 species in subtropical and tropical regions of Asia and Africa (Xiao *et al.*, 1987, He *et al.*, 2002), 2 species were reported from Western Ghats and Courtallum in Tinnevelly District of Tamil Nadu state (Gamble, 1986). The International Plant Names Index (IPNI) includes 187 species of *Tricalysia* (http://www. Plant systematics. org). *Tricalysia A. Rich.* comprises of about 10 spp. in Tropical Africa, Madagascar and few in Indomalaya (George Usher, 1984). It is also found in central and south Maharashtra Sahyadris (Almeida, 2001). *Tricalysia sphaerocarpa* (Dalzell ex Hook. F.) Gamble is not recorded from the tropical dry evergreen forest. This species is known only from Western Ghats and its occurrence is uncommon for the entire east coast and could be considered to be the relict of the past wetter regimes of the Cuddalore district (Israel Oliver King, 2004).

Parthasarathy *et al.*, (2008) reported that *Tricalysia sphaerocarpa* and *Lepisanthes tetraphylla* are the dominant evergreen trees in Thirumanikutzi sacred grove (Cuddalore, Tamil Nadu). *Tricalysia sphaerocarpa* is the most abundant species in Kuzhanthaikuppam (Cuddalore, Tamil Nadu), probably due to past disturbance (Mani and Parthasarathy, 2006). The wild coffee *Tricalysia sphaerocarpa* contributed 50 % of multistemmed individuals in Arasadikuppam and the site was dominated by 33% of the stand (Venkateswaran and Parthasarathy, 2003). Anbarashan and Parthasarathy (2013) have reported that *Tricalysia sphaerocarpa* formed 72% of the forest stand density at S.Pudhoor.

The studies of Mike O. Soladoye *et al.*, (2010) on ethnobotanical survey of anti-cancer plants in Ogun state, Nigeria, revealed that the bark of *Tricalysia*
*macrophylla* along with some other plants and the fruit juice of *Citrus medica* is used to cure cancer. Chris Long (2005) studied the ethnobotanical uses of *Tricalysia capensis* and *Tricalysia lanceolata*. Moshi *et al.*, (2009) studied the ethnobotanical uses of *Tricalysia coriacea* and *Tricalysia coriacea* sbsp. *Nyassae*. George usher (1984) studied the ethnobotanical uses of *Tricalysia sphaerocarpa*. Prajapat and Kumar (2005) studied the ethnobotanical uses of *Tricalysia sphaerocarpa* and *Tricalysia singularis*.

The bioassay-guided fractionation scheme identified the triterpenoids ursolic and oleanolic acids from *Tricalysia niamniamensis* Hiern, demonstrated DNA ligase inhibition profiles to other triterpenes such as aleuritolic acid. Protolichesterinic acid, swertifrancheside and fulvoplumierin represent three additional natural-product structural classes that inhibit hLI (human Ligase I). Fagaronine chloride and certain flavonoids are also among the pure natural products that were found to disrupt the activity of the enzyme, consistent with their nucleic acid intercalative properties. Further analysis revealed the step of the ligation reaction, indicating a direct interaction with the enzyme protein (Tan *et al.*, 1996).

He *et al.*, (2002) isolated seven rearranged ent-Kaurane glycosides, named tricalysiosides A-G(1-7) from the leaves of *Tricalysia dubia* collected from Okinawa island. Their C-18 and 19 methyls were found to have rearranged to form and alpha, beta- unsaturated gamma-lactone ring, with other functional groups remotely located only on C-15,-16, and -17 of the five membered ring. Using X-ray crystallographic analysis, the structure of tricalysioside A(1) was
determined. On the basis of the crystal structure of 1, the structures of the other tricalysiosides (2-7) were also established.

He et al., (2005) isolated eight ent-kaurane glucoside from the leaves of *Tricalysia dubia*. The structure of tricalysiose H (1) was established by X-ray crystallography and those of tricalysiosides I-O (2-8) were elucidated by analysis of spectroscopic evidence.

Four rearranged ent-kaurane diterpenoid alkaloids, tricalysiamides A-D (1-4) having a cafestol-type carbon framework were isolated from the wood of *Tricalysia dubia*. Their absolute structures were determined on the basis of 2D NMR spectroscopy, X-ray crystallographic analysis and chemical methods (Nishimura et al., 2007).

Tamaki et al., (2008) isolated 2 new rearranged ent-kaurane derivatives namely tricalysiolides H and I from the EtOAc-soluble fraction of an MeOH extract of the stem of *Tricalysia dubia*, together with 5 known rearranged ent-kauranes, i.e. tricalysiolides A-E, stigmast-4-en-6beta-ol-3-one, (+)-pinoresinol, scopoletin and syringaldehyde. Their structure were elucidated from the spectroscopic evidence, and their cytotoxicity toward KB cells and P-gp inhibitory activity were assayed.

Shitomoto et al., (2010) isolated one new megastigmane gentiobioside namely tricalysionoside A (1) and 3 sulfates, named sulfatricalysines A-C (2-4) from the water-soluble fraction of a MeOH extract. Sulfatricalysines D-F(5-7) and 3 new ent-kaurane glucosides namely tricalysiosides X-Z(8-10) from the 1-BuOH–soluble fraction of a MeOH extract of leaves of *Tricalysia dubia*. 
Xu et al., (2010) isolated two new ent-kaurane glycosides namely tricalysiosides V and W, with an acylated diasaccharide moiety at the C-3 position from the roots of Tricalysia okelensis and their structures established by spectroscopic and chemical methods.

The studies of Anandhi and Pragasam (2013a) on pharmacognostical and preliminary phytochemical studies on leaf extracts of Tricalysia sphaerocarpa revealed the marked presence of carbohydrate, glycosides, alkaloids, tannin, flavanoids, moderate presence of protein, phenol, terpenoids and saponin and absence of triterpenoids, anthraquiones, catechins, coumarins. Anandhi and Pragasam (2013b) identified 17 phytochemicals from the stem of Tricalysia sphaerocarpa through GC-MS analysis. They have concluded that the plant is highly valuable in medicinal usage for the treatment of various human ailments along with the chemical constituents present in it. The compounds need further research on toxicological aspects to develop safe drug. Anandhi et al., (2014) identified 30 phytochemicals from the methanolic extract of leaves of Tricalysia sphaerocarpa through GC-MS analysis among which fatty acid was the major group consists of 9 compounds. Eicosanoic acid was found to be present as the major compound with peak area 35.77% and retention time 21.865 min, followed by octadecanoic acid (18.81%).

2.2. Objectives of the Present Study

Keeping the view of significances of traditional medicine in the field of plant-based drug discovery, the important Indian medicinal plant, Tricalysia sphaerocarpa was selected to carry out the following tasks.
1. **Pharmacognosy**
   - Anatomical studies
   - Histochemical localization
   - Fluorescence analysis

2. **Phytochemistry**
   - Physico-chemical parameters of various parts
   - Successive extractive and Batch extractive values
   - Ph Determination of aqueous extract
   - Preliminary Phytochemical Screening
   - GC-MS analysis of methanolic extract of leaf, stem, root and fruit

3. **Pharmacology**
   - *In-vitro* Antioxidant activity
   - *In-vivo* studies such as Acute toxicity, Anti-depressant activity and Anti-diabetic activities to develop new plant-based drug that may lead to therapeutic significance.