Review of Literature
2.1 ESSENTIAL OILS

Essential oils, volatile oils or aromatic oils, as their name implies are the volatile, odorous principles of plant and animal sources. As they evaporate when exposed to air at ordinary temperatures, they are also called as ethereal oils. They represent essence or active constituent of plant, hence they are also known as essential oils.

Volatile oils are soluble in alcohol, ether and other lipid solvents and practically insoluble in water. They are usually lighter than water, possess characteristic odours and have refractive indices. Most of them are optically active. They are secreted in special structures such as duct, cell, schizogenous or lysigenous glands, trichomes etc. They are commonly found in the species of families Lamiaceae, Rutaceae, Piperaceae, Zingiberaceae, Apiaceae, Myrtaceae and Lauraceae (Kokate et al., 2005).

2.1.1 Composition of essential oils

Volatile oils are generally mixtures of hydrocarbons and oxygenated compounds derived from these hydrocarbons. In some oils, the hydrocarbons predominate and only limited amounts of oxygenated constituents are present; in others the bulk of the oil consists of oxygenated compounds. The odour and taste of volatile oils is mainly determined by these oxygenated constituents, which are to some extent soluble in water but more soluble in alcohol. Many oils are terpenoid in origin, a smaller number such as those of cinnamon and clove contain principally aromatic (benzene) derivatives mixed with the terpenes. A few compounds, although aromatic in structure, are terpenoid in origin. Volatile oils differ from fixed oils in various ways. They evaporate at room temperature, and can be distilled from their natural sources. They are not glyceryl esters of fatty acids and therefore, cannot be saponified with alkalis (Evans, 2002).

2.1.2 Isolation of essential oils

Essential oils are isolated by following methods:
2.1.2.1 Distillation

The process of converting liquid into vapour and again condensing to liquid is called distillation. The method of distillation depends on the condition of plant material. There are three methods used for oil distillation:

a) **Water distillation**: This process is used in those plant materials, which do not get destroyed by boiling. The material is boiled with water, vapors are condensed and volatile oil is separated. Isolation of turpentine oil is the main example of this method.

b) **Water and steam distillation**: In this process dry and fresh plant material is used, which may be injured by boiling. The material is grounded and dipped in water. A stream of steam is passed in macerated material. The oily layers condense and distillate is separated, e.g., cinnamon oil, clove oil.

c) **Direct steam distillation**: This process is used for fresh plant materials, which contain moisture in considerable amount, so there is no need of maceration. Fresh plant materials like peppermint, spearmint are placed in perforated basket or tray. The steam under pressure is passed through fresh plant material and volatile oil is collected with condensed vapors.

2.1.2.2 Expression:

Some volatile oils are obtained by expression because they cannot be obtained by distillation process without decomposition.

a) **Sponge Method**: Most citrus essences are extracted by means of expression, and in the past were done by hand where the fruit pulp was removed with the rind and pith, then soaked in warm water to make the rind more pliable, since the pith of the fruit absorbed the water. After the fruit has absorbed the water and become more elastic, it was inverted which helped to rupture the oil cells and a sponge placed next to the rind. It was then squeezed to release the volatile oil, which was then collected directly into the sponge. As soon as the sponge
became saturated with oil, it was squeezed and the essential oil collected in a vessel and then decanted.

b) **Ecuelle method:** This form of expression is used mainly to obtain citrus essential oils, and is a little less labour intensive than that of the sponge method. This is a more modern way of essential oil extraction and is referred to as the “ecuelle a piquer process” (direct translation = basin, to prick/stick/prod) where the fruit is placed in a device and rotated with spikes on the side puncturing the oil cells in the skin of the fruit. This cause the oil cells to rupture and the essential oil, and other material such as pigment, to run down to the center of the device which contains a collection area. The liquid is thereafter separated and the oil is removed from the water-based parts of the mixture and decanted.

### 2.1.2.3 Extraction method

The volatile oil is obtained by solvent – solvent extraction with the help of organic liquid like petroleum ether and benzene. This process is performed at 50° C, so that maximum volatile oil is collected. It is a significant process for perfumery industry because it contains more natural odour than the oil obtained by distillation. This process is economical as compared to distillation process in perfumery industry.

### 2.1.2.4 Enfleurage method

Enfleurage is a process that uses odourless fats that are solid at room temperature to capture the fragrant compounds exuded by plants. The fixed oil or a fat is smeared on the glass sheet and fresh flowers containing volatile oil are spreaded on it. The flowers are removed from smeared fat with hand. Volatile oil is absorbed by fat or fixed oil and then extracted using alcohol. This process is used in perfumery industry (Singh and Bhandari, 2008).

### 2.1.3 Therapeutic uses of essential oils

Many essential oils are used as carminative, relaxing the gastric sphincter and encouraging eructation (belching) of the stomach. Further down the gut, the effect typically is antispasmodic. Typical ingredients for such applications include eucalyptus oils, menthol, capsaicin, anise and camphor. Some essential oils work well for upper
respiratory tract and bronchial problems as mild expectorants and decongestants. Some act as locally anaesthetic counterirritants, and thereby exert an antitussive effect.

Some essential oils, such as those of juniper and agathosma are valued for their diuretic effects. With relatively recent concerns about the overuse of antibacterial agents, many essential oils have seen resurgence in off-label use for such properties and are being examined for this use clinically.

Many essential oils affect the skin and mucous membrane. They are used in antiseptics and liniments in particular. Typically, they produce rubefacient irritation at first, and then counterirritant numbness. Turpentine oil and camphor are two typical examples of oils that cause such effects. Menthol and some others produce a feeling of cold followed by a sense of burning. This is caused by its effect on heat sensing nerve endings. Some essential oils, such as clove oil or eugenol, were popular for many years in dentistry as antiseptics and local anaesthetics. Thymol also is well known for its antiseptic effects.

Essential oils often have an odour and are therefore used in food flavoring and perfumery primarily beverages, candies, cosmetics, soaps, candles, mouthwashes, toothpastes etc (Kalia, 2005).

2.2 LITERATURE REVIEW OF CINNAMOMUM ZEYLANICUM BLUME

Cinnamomum zeylanicum Blume, the evergreen tree of tropical area, is considered to be the native of Sri Lanka and Malabar Coast of India and up to a limited extent in eastern India (Kokate et al., 2009).

2.2.1 Taxonomical classification

<table>
<thead>
<tr>
<th>Domain</th>
<th>Eukaryote</th>
</tr>
</thead>
<tbody>
<tr>
<td>Kingdom</td>
<td>Plantae</td>
</tr>
<tr>
<td>Subkingdom</td>
<td>Viridaeplantae</td>
</tr>
<tr>
<td>Phylum</td>
<td>Magnoliophyta</td>
</tr>
<tr>
<td>Division</td>
<td>Mangoliopsida</td>
</tr>
<tr>
<td>Order</td>
<td>Laurales</td>
</tr>
<tr>
<td>Family</td>
<td>Lauraceae</td>
</tr>
<tr>
<td>Genus</td>
<td>Cinnamomum</td>
</tr>
</tbody>
</table>
Species - Zeylanicum

2.2.2 Synonyms

<table>
<thead>
<tr>
<th>Language</th>
<th>Synonym</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sanskrit</td>
<td>Tamalpatra</td>
</tr>
<tr>
<td>Hindi</td>
<td>Dalchini</td>
</tr>
<tr>
<td>Bengali</td>
<td>Dalchini</td>
</tr>
<tr>
<td>English</td>
<td>Cinnamon</td>
</tr>
<tr>
<td>Gujarati</td>
<td>Dalchini</td>
</tr>
<tr>
<td>Tamil</td>
<td>Cannalavangapattai</td>
</tr>
<tr>
<td>Malayalam</td>
<td>Kulit-manis</td>
</tr>
<tr>
<td>Marathi</td>
<td>Dalchini</td>
</tr>
<tr>
<td>Telugu</td>
<td>Dasamchakkalu</td>
</tr>
<tr>
<td>Urdu</td>
<td>Darchini</td>
</tr>
<tr>
<td>Uriya</td>
<td>Dalochini (Nadkarni, 2010)</td>
</tr>
</tbody>
</table>

2.2.3 Botanical description

It is a moderate sized tree, upto 16 m in height. Leaves opposite or sub opposite, glabrous, thinly to stiffly coriaceous, oval or elliptic to lanceolate; flowers yellowish green in axillary panicles; fruits ellipsoid to oblong-ovoid, dark purple, upto 12.5 mm long, Figure 11. The bark of the tree is the well-known Ceylon cinnamon of commerce. In India cultivation of cinnamon is comparatively of recent times, though in the wild form it occurs in the evergreen forests of Western Ghats (The Wealth of India, 1992). The British Pharmacopoeia, (1980) specifies cinnamon as the dried bark of the shoots of coppiced trees of *Cinnamomum zeylanicum*, freed from the outer cork and the underlying parenchyma. The outer surface is dull yellow to brown, fragrant, found in the form of compound quills; taste is aromatic and sweet followed by warm sensation. The outer surface of the bark is marked by wavy longitudinal striations with small holes of scars left by the branches. The inner surface also shows the longitudinal striations, Figure 12, (Wallis, 2005).

Dutta and Dutta, (1955) had described the pharmacognostic characters and distinguished features of the commercial samples of cinnamon bark sold in the Indian market.
Fig. 11: *Cinnamomum zeylanicum* Blume

Fig. 12: *Bark of Cinnamomum zeylanicum* Blume
2.2.4 Description of drug

The drug consists of volatile oil of the bark of *Cinnamomum zeylanicum* Blume (Lauraceae). The oil is obtained by steam distillation of the bark of *Cinnamomum zeylanicum*. It is clear, light yellow with characteristic odour reminiscent of cinnamic aldehyde. The oil has relative density 1.000 to 1.030, refractive index 1.572 to 1.591, optical rotation -2° to +1° (European Pharmacopoeia, 2010).

2.2.5 Medicinal properties and uses

Bark is carminative, antispasmodic, aromatic, stimulant, haemostatic, antiseptic, stomachic and germicide (Satyavati *et al*., 1976). It is used as cordial in cramps of stomach in syncope, in paralysis of tongue and to block the nerve in toothache. It is also used as an aromatic to mask the disagreeable taste of other drugs (I.P, 1966). The oil possesses antibacterial and antifungal properties (Bruneton, 1999). The oil is styptic, emmenagogue; tonic to the liver, useful in inflammation, vomiting and abdominal pains (Kirtikar and Basu, 1984). The oil is a valuable flavouring ingredient used widely in all kinds of confectionary, baked foods, meat seasonings, candies, soft drinks, ketchups, pickles, sauces, beverages, pharmaceutical and dental preparations, mouth rinses etc (Heber, 1950).

2.2.5.1 Phytochemistry

Paranagama *et al.* (2001) had studied essential oils of bark, leaf, root and fruit of *C. zeylanicum* by GC/MS. At least 37 compounds were detected in cinnamon bark oil, out of which 35 were identified. Most of the constituents of the essential oil are terpenes, consisting of monoterpene, sesquiterpene and phenyl proponoids. The oil was found to consist of cinnamaldehyde (50.5%), cinnamyl acetate (8.75%), β- caryophyllene (8.0%), 1,8-cineole (4.60%), eugenol (4.15%), benzyl benzoate (1.10%), α- humulene (1.3%). The other minor components reported in the study were cinnamyl alcohol, 2-phenyl ethyl alcohol, benzaldehyde, hydrocinnamaldehyde, 2 phenyl ethyl acetate, methyl cinnamate, eugenyl acetate, isoeugenol, safrole, and α- ylangene.

The chemical composition of bark oil was also studied by Senanayake *et al.* (1978). They also reported cinnamaldehyde, cinnamyl acetate, β- caryophyllene, 1, 8- cineole, eugenol
and benzyl benzoate as major components of the oil. The structures of some of the important constituents are shown in Figure 13-23.

Fig.13: Cinnamyldehdye

Fig.14: Cinnamyl acetate

Fig.15: β-caryophyllene

Fig.16: 1,8- Cineole

Fig.17: Eugenol

Fig.18: Benzyl benzoate

Fig.19: α-humelene

Fig.20: Cinnamyl alcohol

Fig.21: 2- Phenylethyl alcohol

Fig.22: Hydrocinnamaldehyde
2.2.6 Pharmacological activities

2.2.6.1 Antibacterial activity

The oil from the bark exhibits potent antimicrobial activity (Chang et al., 2001 and DE et al., 1991). A study was conducted to isolate the most bioactive compound from the bark oil of *C. zeylanicum*. The isolated material was investigated for its antibacterial activity against six selected bacteria. Cinnamaldehyde at different concentration was active against all the tested bacteria and the highest inhibitory effect was observed against *Bacillus cereus* using disc diffusion method (Al-Bayati and Mohammed, 2009).

Shahverdi et al. (2007) reported that the oil of *C. zeylanicum* bark enhanced the bactericidal activity of clindamycin and also decreased the MIC of clindamycin required for toxigenic strain of *Clostridium difficile*. A trans-cinnamaldehyde fraction was isolated from the oil and it showed synergistic actions of clindamycin 16 fold at concentration 20 μg/ml. The results signified that low concentration of trans cinnamaldehyde elevate the antimicrobial action of clindamycin.

*Escherichia coli* is a pathogenic strain that causes hemorrhagic colitis, hemolytic uremic syndrome and thrombocytopenic purpura in humans. The control of bacterial cells in food is an important factor to reduce food borne diseases due to *E. coli* 0157: H7. Assays to inactivate *E. coli* were carried out by using cinnamon oil. A dramatic decrease was observed in the viable counts. In the presence of 0.05% of the oil, most of the cells were killed after 30 minutes Senhaji et al., 2007).

2.2.6.2 Antifungal activity

Pandey et al. (2010) reported the antifungal and antioxidant activities of various bioactive fractions extracted from the bark oil and leaves of *C. zeylanicum*. The fungicidal activity of the oil was evaluated against pathogenic fungi namely *Aspergillus flavus*, *A. fumigatus*, *A. niger*, *Pencillium spp* and *Candida albicans*. The antioxidant activities of the fractions were evaluated by using reducing power assay. The results showed the fungicidal and antioxidant effects which may be attributed to the presence of phenolic and flavanoid compounds.
2.2.6.3 Antiviral activity

Premnathan et al. (2000) in their study had screened 69 plant species against HIV-1 and HIV-2. The most effective extracts against the virus were found to be *Cinnamomum zeylanicum* and *Cardiospermum helicacabum*.

Wen et al. (2007) reported that terpenoids present in cinnamon bark oil have potent antiviral property against HSV-1 and HSV-2 (oral and genital herpes) viruses.

In another study *Cinnamomum zeylanicum* bark oil, a component of Japanese medicine Mao-to had showed to have an antiviral therapeutic effect (Wondrak et al., 2010).

2.2.6.4 Antiparasitic activity

The toxicity of cinnamon oil against eggs and adult females of human head louse, *Pediculus humanus capitis* was examined using direct contact and vapour phase bioassays and compared with two widely used pediculicides, d-phenothrin and pyrethrum. Cinnamon oil exhibited good activity against the parasites (Yang et al., 2005).

2.2.6.5 Antidiabetic activity

Interest in cinnamon as a potentially useful treatment for type 2 diabetes began almost 20 years ago. Khan et al. (1990) had isolated an unidentified factor from cinnamon oil and termed it as Insulin Potentiating Factor (IPF). They demonstrated that IPF might be involved in the alleviation of the signs and symptoms of diabetes and other diseases related to insulin resistance.

A study was designed by Rekha et al. (2010) to evaluate the ameliorative effect of *C. zeylanicum* oil upon early stage diabetic nephropathy owing to its antioxidant and antidiabetic effects. The results confirmed a significant protection against diabetic nephropathy.

Broadhurst et al. (2000) compared 49 herbs and medicinal plant extracts for their insulin potentiating action in an *in vitro* model. The aqueous extract of *C. zeylanicum* potentiated insulin activity more than 20 folds, higher than any other compound.
2.2.6.6 Antitumor activity

It is suggested that the cinnamon derived dietary factor cinnamic aldehyde activates the Nrf 2-dependent antioxidant response in human epithelial colon cells and may therefore represent an experimental chemopreventive dietary factor targeting colorectal carcinogenesis (Wondrak et al., 2010). Anti melanoma activity of cinnamic aldehyde was observed in cell culture and mouse model of human melanoma (Orihara et al., 2008).

Golamreza et al. (2010) investigated tumor inhibition activity of essential oil of C. zeylanicum. It was found that the oil had inhibition effects on Agrobacterium tumefaciens that induced crown gall tumor.

2.3 REVIEW OF LITERATURE OF PONGAMIA GLABRA VENT.

A medium sized glabrous tree, upto 18 m high, found almost throughout India up to an altitude of 1,200 m and further eastwards (Khare, 2004).

2.3.1 Taxonomical classification

<table>
<thead>
<tr>
<th>Kingdom</th>
<th>Plantae</th>
</tr>
</thead>
<tbody>
<tr>
<td>Division</td>
<td>Magnoliophyta</td>
</tr>
<tr>
<td>Class</td>
<td>Magnoliopsida</td>
</tr>
<tr>
<td>Order</td>
<td>Fabales</td>
</tr>
<tr>
<td>Family</td>
<td>Papillionaceae</td>
</tr>
<tr>
<td>Genus</td>
<td>Pongamia</td>
</tr>
<tr>
<td>Species</td>
<td>Glabra</td>
</tr>
</tbody>
</table>

2.3.2 Synonyms

<table>
<thead>
<tr>
<th>Bengali</th>
<th>Karanj</th>
</tr>
</thead>
<tbody>
<tr>
<td>English</td>
<td>Pongam oil tree</td>
</tr>
<tr>
<td>Hindi</td>
<td>Karanj</td>
</tr>
<tr>
<td>Gujrati</td>
<td>Karanja</td>
</tr>
<tr>
<td>Kannada</td>
<td>Honge</td>
</tr>
<tr>
<td>Marathi</td>
<td>Kidamar</td>
</tr>
<tr>
<td>Tamil</td>
<td>Ponga, Pungammaram</td>
</tr>
</tbody>
</table>
Telugu - Kanuga, Punagu  
Punjabi - Sukhchein, Karanj

2.3.3 Botanical description of the plant

It is a medium sized evergreen tree with a spreading crown and a short bole. The tree is planted for shade and is grown as ornamental tree. It is one of the nitrogen fixing trees producing nitrogen. It is fast growing, glabrous, deciduous, trunk diameter upto 60 cm, bark smooth, grey; leaves imparipinnate, shiny, young, pinkish red, mature leaves are glossy and deep green; leaflets- 5-9, the terminal leaflet is larger than the others; flowers-fragrant white to pinkish, paired along rachis in axillary pendent, long racemes or panicles cup shaped, truncate; stamens-monadelphous vexillary; ovary subsessile to short stalked, pubescent, ovules two, glabrous, stigma small and terminal; pod short stalked, oblique oblong, flat, smooth, thickly leathery to subwoody, indehiscent, one seeded, Figure 24 and 25 (Chopde et al., 2008).

![Pongamia glabra Vent.](image)

Fig.24: *Pongamia glabra* Vent.
2.3.4 **Description of the drug**

The drug consists of fixed oil obtained from the seeds of *Pongamia glabra* Vent. (Papillionaceae), Syn. *Pongamia pinnata*. Seeds are one and rarely two per pod, elliptic or reniform in shape, 1.7- 2.0 cm long and 1.2- 1.8 cm broad, wrinkled with reddish leathery testa, microphylar end of cotyledons slightly depressed while other side semi-circular in shape. Israeli and Issar, (1977) had described the pharmacognostic characters of seeds of *P. glabra*. The oil is extracted from the seeds by cold press process method. The oil has specific gravity 0.91 to 0.940, refractive index 1.47 to 1.4790, acid value 20, saponification value 186 to 196, and unsaponifiable matter 3.0.

2.3.5 **Medicinal properties and uses**

The seeds are used for external application in skin diseases. The oil from the seeds is used in leucoderma, cutaneous infections including herpes and scabies, and also in rheumatism. Internally the oil is used as a stomachic, cholagogue and in dyspepsia with sluggish liver. The seeds crushed into a paste are used externally for skin diseases including leprous sores and also as a fish poison. The powdered seeds are considered to be good expectorant in bronchitis and whooping cough. The seed oil is antibacterial and antifungal (Medicinal Plants of India, 1987). The oil is source of biodiesel and is used as fuel for cooking and lamps (Mahli *et al.*, 1989).

2.3.6 **Phytochemistry**
A number of compounds have been isolated and characterized from *P. glabra* and the industrial use of some of the compounds is established. The chemical constituents and uses of the plant have been reviewed by Parmar *et al.* (1976).

Karanjin, the principal furanoflavonoid constituent was isolated from the seed oil (Limaye, 1925). The other furanoflavonoids identified and characterized in the seed oil were pongapin (Aneja *et al.*, 1958), pongaglabrone (Khanna and Seshadri, 1963). *P. glabra* yielded two chromeno-flavones viz., isolonchocarpin (Naik and Bringi, 1973a) and karanjachromene (Naik and Bringi, 1973b).

The seed oil of the drug yielded two furanodiketones, viz., pongamol and ovalitenone (Rangaswami and Seshadri, 1942; Narayanaswami *et al.*, 1954; Mukeerjee and Seshadri, 1956). The seed oil also yielded a simple flavone, desmethoxykanjugin (Mittal and Seshadri, 1956). The oil yielded known compounds like karanjin, pongapin, lanceolatin B, kanjone and isopongaflavone (Roy *et al.*, 1977).

The seed oil of *P. glabra* yielded number of fatty acids like oleic and linoleic acids and beta sitosterol (Badami and Daulatabad, 1967; Sinha, 1959). The structures of some of the important constituents are shown in Figure 26-35.

![Fig.26: Karanjin](image26.png)

![Fig.27: Pongapin](image27.png)

![Fig.28: Pongalborne](image28.png)

![Fig.29: Pongamol](image29.png)

![Fig.30: Karanjachromene](image30.png)

![Fig.31: Ovalitenone](image31.png)
2.3.7 Pharmacological activities

2.3.7.1 Antimicrobial activity

The seed oil showed antibacterial activity against *Micrococcus pyogenes* var. *aureus*, *Mic. Pyogenes* var. *citreus*, *Bacillus subtilis*, *Salmonella typhosa*, *Sal. paratyphi A*, and *Sal. paratyphi B* and *Escherichia coli*. The oil was found to be more active than neem oil (Patel and Trivedi, 1962).

Karanjin showed good antibacterial activity against *Mycobacterium tuberculosis*, and inhibits the growth of the organism at a concentration of 10 ppm (Ramaswami and Sirsi, 1967).

Jambotkar *et al.* (1962) evaluated soaps prepared from some inedible oils for antibacterial activity against *Aspergillus niger* and *Trichophyton gypseum*.

A study by Wagh *et al.* (2007) revealed the antifungal and antibacterial activity of seed oil of *P. glabra*. The results concluded very significant antymycotic activity against *Aspergillus niger* and *A. fumigatus*.

Bawa *et al.* (2001) evaluated the antibacterial activity of karanj oil *in vitro* against 14 strains of pathogenic bacteria using the tube dilution technique. The activity of the oil was bactericidal and independent of temperature and energy. Most of the pathogens were killed at 4°C. The activity was mainly due to the inhibition of cell membrane synthesis in the bacteria.
2.3.7.2 Sunscreen activity
The sunrise protective factor (SPF) property of pongamol from seeds of *P. pinnata* in the U.V. region and its effects were compared with well established standard sunscreen drugs P-amino benzoic acid (PABA) and Avobenzene. The seed extract was found to be highly effective sunscreen in UVA region as shown by the drug Avobenzene where as PABA showed its protective action in the UVB and UVC regions (Buddepu et al., 2011).

2.3.7.3 Larvicidal activity
The activity of water extract of the oil seed cake of *P. glabra* was reported against second stage larvae of *Meloidogyne incognita* (Mishra and Prasad, 1973).

The larvicidal property of seed oil was evaluated in combination with neem oil. The combination of both the oils in equal proportion proved to have better activity against mosquitoes, *Culex quinquefasciatus*, *Anopheles stephensi* and *Aedes aegypti* (Govindrajan, 2008).

2.3.7.3 Antiviral activity
The seed extract of *P. glabra* was evaluated against HSV-1 and HSV-2 *in vitro*. The most striking observation was the total inhibition of growth of HSV-1 and HSV-2 at concentration of 1 mg/ml and 20 mg/ml w/v respectively (Singh et al., 1996).

2.3.7.4 Antidiabetic activity
*P. glabra* is reported to have shown hypoglycemic effect in normal and alloxan diabetic rabbits (Aiman, 1970).

2.3.7.6 Spermicidal activity
Bandivdekar and Moodbidri (2002) investigated the spermicidal activity of seed oil and found that it exhibits strong spermicidal activity.

2.4 REVIEW OF LITERATURE OF EUCALYPTUS GLOBULUS LABILL.
It is a native of Australia now being cultivated on the highlands of India, chiefly on the Nilgiri, Malabar and Coorg. It is also planted in plains of North India and in the mountainous tracts (Satyavati et al., 1976).

2.4.1 Taxonomical classification
Kingdom - Plantae
Subkingdom - Tracheobionta
Division - Magnoliophyta
Class - Magnoliopsida
Order - Myrtales
Family - Myrtaceae
Genus - Eucalyptus
Species - Globulus

2.4.2 Synonyms
English - Blue gum tree
Hindi - Yukeliptus, sapheda
Sanskrit - Tailaparna
Malayalam - Yukalimaram
Marathi - Nilgiri
Tamil - Yukkaalimaram
Telugu - Jeevakamu (The Ayurvedic Pharmacopoeia of India, 2004)

2.4.3 Botanical description
In *Eucalyptus globulus*, the leaves are 20-25 cm long, bifacial, lanceolate, scythe-shaped, glabrous, sessile, thin and wax coated, **Figure 36-37**. Microscopically there is a coating of wax over and above the thin cuticle, ranunculaceous type stomata are present on lower surface only, few round internal oil glands are present and calcium oxalate crystals of prismatic and rosette type are also seen (Datta and Datta, 1952).
2.4.4 Description of drug
The drug consists of volatile oil obtained from the leaves of *Eucalyptus globulus* Labill. (Myrtaceae). The oil is colourless or pale yellow liquid that has characteristic, aromatic somewhat camphoraceous odour and a pungent, spicy and cooling taste. It exhibits optical rotation 0° to +10°, refractive index 1.457-1.469, weight per ml 0.897-0.924 g (I.P. 2007 and Tyler *et al.*, 1981).

### 2.4.5 Medicinal properties and uses
Leaves are febrifuge and carminative, stimulant, expectorant, diaphoretic and antiseptic. Eucalyptus oil is a powerful antiseptic and disinfectant, antimalarial, rubefacient, stimulant, antispasmodic and it is much used as an inhalant. It is widely used in curing headache and body pains. Indian Pharmaceutical industry is using the oil largely as a mosquito repellent and as an ingredient of germicidal and disinfecting preparations. It is used as an antiseptic especially in the treatment of infections of upper respiratory tract and in certain skin diseases. It is found useful in rheumatism and in chronic bronchitis and asthma (Kumar, 1988 and Schnitzler *et al.*, 2001).

### 2.4.6 Phytochemistry
Eucalyptus oil contains over 80% 1,8- cineol, p-cymene (2.7%), alpha-pinene (2.6%), limonene (0.5%), geraniol, camphene, alpha phellandrene and euglobal. The structures of the important constituents are shown in Figure 38-45.

Fig.38: 1,8 – Cineole

Fig.39: p-Cymene

Fig.40: α-Pinene

Fig.41: Limonene
2.4.7 Pharmacological activities

2.4.7.1 Antimicrobial activity

Noumi et al. (2010) had evaluated essential oil of *E. globulus* for its ability to inhibit the mycelium formed on Lee medium by oral *Candida albicans* strains. The result obtained showed that the oil inhibit total mycelium in *C. albicans* isolate at MIC 0.312 mg/ml. Vilela et al. (2009) had evaluated the effectiveness of essential oil from leaves of *E. globulus* against the fungi *Aspergillus flavus* and *A. parasiticus*. It was found that complete fungal growth inhibition of both species was achieved with the essential oil.

Ghalem et al. (2008) had determined the antibacterial activity of essential oil of *E. globulus* and *E. camaldulensis* against *Staphylococcus aureus* gram (+) and *Escherichia coli* gram (-) bacteria. Results demonstrated that both the species of eucalyptus showed an excellent inhibitory effect on *S. aureus* than that of *E. coli*. Authors suggested the potential usefulness of two *Eucalyptus* species as a microbiostatic, antiseptic or as a disinfectant
agent. Chhetri et al. (2008) had also reported the excellent activity of oil of _Eucalyptus globulus_ against _S. aureus_ and _E. coli_.

In another study, the antimicrobial activity of _Eucalyptus globulus_ essential oil was evaluated against 14 food spoilage microorganisms in liquid and vapour phase. The MIC varied from 2.25 to 9mg/ml for bacterial and fungal strains and from 1.13 to 2.25mg/ml for yeast strains. Significantly higher antimicrobial activity was observed in the vapour phase (Tyagi and Malik, 2011).

### 2.4.7.2 Analgesic and anti-inflammatory activity

Analgesic and anti-inflammatory activity of the oil is well established. _E. globulus_ oil brings out pain relief by its counter irritant, anti-inflammatory and nociceptive actions. Silva et al. (2003) evaluated the analgesic and anti-inflammatory effects of essential oil extract of three species of eucalyptus namely _Eucalyptus globulus_, _E. citriodora_ and _E. tereticornis_. The result of the study suggested that essential oil extracts of all the three species of eucalyptus possess central and peripheral analgesic effects as well as neutrophil dependent and independent anti-inflammatory activities.

### 2.4.7.3 Insecticidal activity

The insecticidal activity of _E. globulus_ oil against the larva and adult housefly, and also against _Musa domestica_ was evaluated under controlled laboratory condition. At concentrations of 0.5, 0.3, 0.2 and 0.1 % the larval mortality rate was 90 % (Haliman, 2005).

### 2.4.7.4 Anthelminitic activity

Taur et al. (2010) had evaluated the anthelminitic activity of volatile oil isolated from _E. globulus_ on adult Indian earthworms, _Pheretima posthuma_ that has anatomical and physiological resemblance with the intestinal roundworm parasites of human beings. The oil showed potent anthelmintic activity as compared to that of the standard drug albendazole at a concentration of 10 mg/ml.

### 2.4.7.5 Larvicidal activity
E. globulus leaves have potent action against Culex quinquefasciatus and Culex tritaeniorhynchus (Monzan et al., 1994).

2.4.7.6 Antiviral activity

Twelve euglobals from E. globulus and their twenty six related compounds were examined for their inhibitory effects on Epstein- Barr virus activation by a short term in vitro assay. The results showed that most of the euglobals having monoterpenes structures and euglobal-111 had strong inhibitory activity.

E. globulus oil has antiviral activity against Herpes simplex virus 1 and 2 (Schnitzler et al., 2001). In another in vitro study, it was reported that oil of E. globulus has mild but noticeable reduction effect for mumps virus, but no effect on adenovirus (Cermelli et al., 2008).

2.5 REVIEW OF LITERATURE OF OCIMUM KILIMANDSCHARICUM

Ocimum kilimandscharicum is a native of Africa and was introduced and cultivated in India and some parts of Turkey. In India, it is cultivated in West Bengal, Assam, Tamil Nadu, Karnataka, Kerala, Dehradun and in North India (Khare, 2007).

2.5.1 Taxonomical classification

<table>
<thead>
<tr>
<th>Classification</th>
<th>Kingdom</th>
<th>Plantae</th>
</tr>
</thead>
<tbody>
<tr>
<td>Domain</td>
<td>Eukaryota</td>
<td></td>
</tr>
<tr>
<td>Kingdom</td>
<td>Plantae</td>
<td></td>
</tr>
<tr>
<td>Sub Kingdom</td>
<td>Tracheobionta</td>
<td></td>
</tr>
<tr>
<td>Division</td>
<td>Spermatophyta</td>
<td></td>
</tr>
<tr>
<td>Class</td>
<td>Magnoliopsida</td>
<td></td>
</tr>
<tr>
<td>Subclass</td>
<td>Asteridae</td>
<td></td>
</tr>
<tr>
<td>Order</td>
<td>Lamiales</td>
<td></td>
</tr>
<tr>
<td>Family</td>
<td>Lamiaceae</td>
<td></td>
</tr>
<tr>
<td>Genus</td>
<td>Ocimum</td>
<td></td>
</tr>
<tr>
<td>Species</td>
<td>Kilimandscharicum</td>
<td></td>
</tr>
</tbody>
</table>

2.5.2 Synonyms
2.5.3 Botanical description of the plant

It is a woody shrub that can reach 2 m high in warm temperature regions of the tropics but can be propagated by seeds and vegetatively. The plant has pubescent quadrangular branchlets with simple leaves that are opposite and oblong narrows at the base and deeply serrated (Warrier et al., 1996). Flowers are white, pedicel straight, calyx ovoid to campanulate, corolla tube slightly shorter than calyx or rarely exerted, stamens 4, anthers ovoid reniform, style longer than stamen. Fruits are one seeded, indehiscent type, found in clusters, tap roots are deep and soft wooded, Figure 46-47. The leaves accommodate aromatic oil, which represents the essence of plant (Saha et al., 2010 and Darrah, 1974).
2.5.4 Description of drug
The drug consists of volatile oil obtained from the leaves of *Ocimum kilimandscharicum* (Lamiaceae). The plant is reported to yield essential oil from leaves 0.77 – 1.12 % (dry wt. basis). The oil is colourless to light yellow liquid that has strong odour of camphor with pungent and camphoraceous taste. It exhibits optical rotation 12 to 14°, refractive index 1.5065 to 1.5450.

### 2.5.5 Medicinal properties and uses

The leaves are acrid, thermogenic, aromatic, insecticidal, antiviral, appetizing, useful in cough and bronchitis, antibacterial and antifungal (Indian Medicinal Plants, 1991). Traditionally it is used in abdominal pains, diarrhoea, congested chest, cough and cold. In Indian System of Medicine (Ayurveda), oil of *Ocimum kilimandscharicum* has been used as an anti-inflammatory, indigestion, insecticidal, mosquito repellent, aromatic and antimicrobial (Kashyap *et al.*, 2011 and Githinji, 1993).

### 2.5.6 Phytochemistry

The plant has been considered to give a promising yield of camphor in India (Chowdhri and Haksar, 1961a, 1964b). The essential oil revealed the presence of camphor (70.43%) limonene (6.23%), pinene (3%), camphene (5.07%), 1,8 cineole (7.20%), linalool (0.47%), 4-terpineol (1.44%), transcaryophyllene, α – selinene, p – cymene and β- phellandrene were also reported in the oil (Chowdhri and Haksar 1962c). Singh *et al.* (2011) and Padalia and Verma, (2011) had also analysed the phytoconstituents of *O. kilimandscharicum* by GC-MS analysis. Apart from the mentioned constituents they also reported the presence of β – myrcene (1.58%), β – ocimene (2%) and ethylamyl carbinol (0.88 %). The structures of some of the important constituents are shown in Figure 48- 58.
Fig. 48: Camphor

Fig. 49: Limonene

Fig. 50: Pinene

Fig. 51: Camphene

Fig. 52: 1,8 – Cineole

Fig. 53: Linalool

Fig. 54: 4-Terpineol

Fig. 55: Transcaryophyllene
2.5.7 Pharmacological activities

2.5.7.1 Antimicrobial activity

Verma et al. (2011) had evaluated oils of *O. kilimandscharicum* and *O. gratissimum* for their chemical composition and antibacterial activity against *Staphylococcus aureus*, *S. epidermidis*, *Enterococcus faecalis* and *Streptococcus mutans*. Both oils showed good activity against tested strain. In another study, the oil was found to be active against *Escherichia coli* and *Pseudomonas aeruginosa* as well as fungus *Candida albicans* (Anand et al., 2011).

2.5.7.2 Antioxidant activity

The total antioxidant activity of oil of *O. kilimandscharicum* was measured in terms of antioxidant index (%) by using two bioassay systems (Chicken liver and muscles). It was observed that even 1.5 ppm concentration of oil is more effective than the standard antioxidant (L-ascorbate). It is evident that *Ocimum kilimandscharicum* showed a significant level of protection against lipid peroxidation from free radical induced damage in both liver and muscle assay systems (Singh et al., 2011).

In another study, Hakkim et al. (2008) had investigated an antioxidative property of eight selected *Ocimum* species including *Ocimum kilimandscharicum* using iron reduction, \( \beta - \text{...} \)
carotene–linoleic acid bleaching and superoxide anion free radical scavenging assays. It was found that *Ocimum* species exhibited activity in all the *in vitro* antioxidant assays. Authors suggested that the phytochemicals in *Ocimum* species are rich antioxidants and can be used as an effective preservative in food industry.

### 2.5.7.3 Insecticidal activity

The essential oils of *O. kilimandscharicum* and *O. suave* were evaluated against malaria vectors in northeastern Tanzania. It was observed and concluded that use of *O. kilimandscharicum* and *O. suave* as repellent would be beneficial in reducing vector biting. Seyoum *et al.* (2003) had also reported the mosquito repellent property of *O. kilimandscharicum* against *Anopheles gambiae sensu lato*.

### 2.5.7.4 Wound healing activity

Paschapur *et al.* (2009) had evaluated the wound healing potential of aqueous extract of leaves of *O. kilimandscharicum*. The activity was examined in three types of wound models on rats: the excision, the incision and dead space wound model. The results of the study showed that the extract of leaves possesses a definite prohealing action. This was demonstrated by a significant increase in the rate of wound contraction and by enhanced epithelization.

### 2.5.7.5 Antitumor activity

Monga *et al.* (2011) evaluated the antimelanoma and radio protective activity of alcoholic aqueous extract of different species of *Ocimum* including *O. ilimandscharicum* in mice. The results showed that 50 % alcoholic extract of *O. Kilimandscharicum* resulted in significant reduction in tumor volume, increase in average body weight and survival rate of mice.

### 2.5.7.6 Acaricidal activity

The essential oil and hydrodistilled extracts of *O. kilimandscharicum* and extracts of *Artemisia annua* and oil seeds of *Pongamia glabra* were tested for their *in vitro* efficiency against *Boophilus microplus*. *O. kilimandscharicum* showed highest efficacy (98.34%) followed by *P. glabra* (96.67%) and *A. annua* (95%) (Vatsya *et al.*, 2006).
2.6 REVIEW OF LITERATURE OF MELALEUCA LEUCADENDRON (L.) L.

Plants are evergreen shrubs or trees found in the East Indies and Australia.

2.6.1 Taxonomical classification

<table>
<thead>
<tr>
<th>Domain</th>
<th>Eukaryota</th>
</tr>
</thead>
<tbody>
<tr>
<td>Kingdom</td>
<td>Plantae</td>
</tr>
<tr>
<td>Division</td>
<td>Magnolia Phyta</td>
</tr>
<tr>
<td>Class</td>
<td>Magnoliopsida</td>
</tr>
<tr>
<td>Subclass</td>
<td>Rosidae</td>
</tr>
<tr>
<td>Order</td>
<td>Myrtales</td>
</tr>
<tr>
<td>Sub order</td>
<td>Myrtleae</td>
</tr>
<tr>
<td>Family</td>
<td>Myrtaceae</td>
</tr>
<tr>
<td>Genus</td>
<td>Melaleuca</td>
</tr>
<tr>
<td>Species</td>
<td>Leucadendron</td>
</tr>
</tbody>
</table>

2.6.2 Synonyms

<table>
<thead>
<tr>
<th>Language</th>
<th>Synonym</th>
</tr>
</thead>
<tbody>
<tr>
<td>English</td>
<td>Cajeput tree</td>
</tr>
<tr>
<td>Hindi</td>
<td>Shitanshu</td>
</tr>
<tr>
<td>Marathi</td>
<td>Vishaha</td>
</tr>
<tr>
<td>Tamil</td>
<td>Kayaputi</td>
</tr>
<tr>
<td>Malayalam</td>
<td>Cajeputier</td>
</tr>
<tr>
<td>Sanskrit</td>
<td>Kayaputi</td>
</tr>
</tbody>
</table>

2.6.3 Botanical description of the plant

The plant grows as shrub or tree with single trunk ranging from 25–40 m in height. The bark is thick and spongy and flakes out in elongated papery pieces from time to time. The branches are pendulous, grow in an irregular ascending manner with dense green foliage. The leaves are thin, narrow and lancet-shaped. It alternates on short footstalks and are shiny green or ash coloured. The leaves are highly aromatic and bitter to taste. The flowers can be recognized by its creamy white elongated spiky petals, spikes long interrupted solitary or 2–3 together terminal at first and then surmounted by leafy branches, rachis and calyx glabrous or wooly. The fruits are usually light brown to dark brown. Seeds are
obovoid or cuneate, very small in size; cotyledons ovate, thick, much longer than the radical (Hooker, 1879 and Tanaman, 2007). Figure 59- 60.

Fig.59: *Melaleuca leucadendron* (L.) L.

Fig.60: Leaves of *Melaleuca leucadendron*
2.6.4 Description of the drug

The drug consists of volatile oil obtained by hydro distillation from the fresh leaves of *M. leucadendron*. The oil distilled from leaves is colourless and limpid with a very pleasant, camphoraceous odour and a bitter aromatic taste (Trease and Evans, 2002). It has specific gravity 0.868 to 0.930, optical rotation - 4° to 0° and refractive index value ranged from 1.4630 to 1.4820. The oil yield ranged from 0.4 to 1.2 % (v/w % fresh weight).

2.6.5 Medicinal properties and uses

The oil of *M. leucadendron* is an antiseptic used externally for thrush, vaginal infections, acne, athlete’s foot, verruca, wart, insect’s bites, cold sore and nits (Thomas, 2000). Traditionally, it is used in rheumatism, stiff joints, neuralgia, migraine, and as mosquito repellent (Khare, 2007). Besides it is used to heal wounds, as topical applications for skin problems such as psoriasis and eczema, used as anthelmintic and parasiticidal agent to treat roundworms, scabies and pediculosis. It is also used to treat cold, fever, influenza, stomach and intestinal problems because of its antibacterial properties (Southwell, 1999; Budiadi, 2005 and Brug, 1947).

2.6.6 Phytochemistry

The volatile oil composition of *M. leucadendron* leaves determined by Gas Chromatography Mass Spectrometry (GC–MS) method revealed the oil to contain terpenoids 1,8 – cineole (40-65%) as major component, with α – pinene, α – terpineol, nerolidol, limonene, benzaldehyde, β– Caryophyllene, valeraldehyde, dipentene and various sesquiterpenes. Other components included l– pinene, terpineol, valeric acid, butyric acid, benzoic acid and aldehydes. The sesquiterpene alcohols, azulene, dipentene, valeraldehyde and benzaldehyde were also found in the volatile oil of leaf and aerial parts. Betulin, firedelin, epitara, xeryl acetate were also found to be present (DeColmenares *et al.*, 1998 and Susanto *et al.*, 2003). The structures of some of the important constituents are shown in Figure 61-65.
Fig. 61: α-Pinene

Fig. 62: Nerolidol

Fig. 63: Valeraldehyde

Fig. 64: Dipentene

Fig. 65: 1,8-Cineole

Fig. 66: Benzaldehyde

Fig. 67: l-Pinene

Fig. 68: Betulene

Fig. 69: Butyric Acid

Fig. 70: Benzoic Acid
Pujiarti et al. (2011) in their study collected the volatile oil of *M. leucadendron* from the leaves of the tree of different ages (5, 10 and 15 years) and studied their chemical composition by GC–MS. The study concluded the presence of 1,8–cineole, α–terpineol, d (+)–limonene and β–caryophyllene as major components in all the samples. Samples from each site tended to decrease in 1,8–cineole content and increase in β–caryophyllene content as plant age increase. 

α–Terpineol was highest at plant age 10 years and d (+)–limonene varied according to plant site and age. 

### 2.6.7 Pharmacological activities

#### 2.6.7.1 Antimicrobial activity

The chemical constituents of *M. leucadendron*, viz, 1,8–cineole, (-)-terpinen 4(ol), (±)α–terpineol and cajeputol have been identified to possess antimicrobial activity. The platyphyllol and similar compounds found in the species that was used in bactericide and fungicide preparations had been patented. The piceatannol a derivative from *M. leucadendron* showed antibacterial activity against four *Helicobacter pylori* strains with MIC values of 25, 50, 12.5, 25 μg/ml respectively (Funatogawa et al., 2004). The oil from *M. leucadendron* was found to be antimicrobial and hyperemic in vitro. It was active against *Bacillus cereus* and *Staphylococcus aureus*, but was inactive against *E.Coli* and *P.aeruginosa* (Decolmenares et al., 1998).

Besides its antibacterial properties, the volatile fraction of the leaves of *Melaleuca leucadendron* has also been reported to contain antifungal activity (Dubey et al., 1983).

#### 2.6.7.2 Anti-inflammatory activity

Ursolic acid, which is a triterpenoid found in *Melaleuca leucadendron* was shown to possess anti-inflammatory property by inhibiting histamine release from mast cell (Liu, 1995).

#### 2.6.7.3 Antioxidant and hepatoprotective activity

Ursolic acid, a steroid like triterpene compound found in *Melaleuca leucadendron* was reported to have strong hepatoprotective activity against ethanol. Ursolic acid showed higher protective action against heart as compared to liver in vitro. The studies involved
induction of ethanol toxicity in wistar rats and ursolic acid was found to control oxidative stress by decreasing lipid peroxidation products and increasing the activities of antioxidant enzyme (Saravanan and Pugalendi, 2006).

2.6.7.4 Anticancer activity
Piceatannol (3 – hydroxyl resveratrol) is a naturally occurring polyphehnl and has been identified as an active component of M. leucadendron. It is a natural analog of resveratrol, a known anticancer agent although it might exhibit a slightly different biological activity (Aggarwal et al., 2004). The studies have shown that this compound was able to inhibit the growth of colorectal cancer cell lines and arrests Caco– 2 cells in the S phase of the cell (Wolter et al., 2002).

2.6.7.5 Insect repellent and pesticidal activity
The oil of M. leucadendron was found to be effective in providing repellency against Aedes aegyti, Anopheles stephensi and Culex quinquefasciatus where it provided a protection time of 8h at the maximum and a 100% repellency against all three species (Abdelkrim and Mehlhorn, 2006). A compound leucadenone A found in M. leucadendron was reported to have a similar structure to a compound that was responsible for the antifeedant nature of Luma chequn (Myrtaceae) an aromatic evergreen shrub, native to Chile while it is immune to insects and pests (Connolly, 2001). M. leucadendron was also analyzed for its antitermite activity (Sakasegawa et al., 2003).