DETERMINATION AND IDENTIFICATION OF THE ESSENTIAL OIL OF VOLATILE OIL EXTRACTED FROM THE EUCALYPTUS LEAF OF THE CITRIODORA SPECIES

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Abstract:
The antifungal bacterial and essential features of essential oils have been reported in many sources around the world which illustrates the potential role of plant bioactive compounds in the various sciences. The identification of the main components of these natural herbal products can be very effective in the pursuit of research in the various fields of science. In this regard, the present research has been carried out to identify and introduce the main components of eucalyptus of the citriodora species. The volatile oil of young and adult eucalyptus leaves was extracted utilizing a Clevenger apparatus and after injection to the extent necessary to the chromatograph gas, using inhibition time and inhibition index, the essential oil composition was studied. From the decomposition of volatile oil of the young and mature eucalyptus leaves of the citriodora, 11 mixtures were extracted. The composition of the citronella in the volatile oil of mature leaves and young leaves were respectively 65.23% and 77.49% as the main composition of the volatile oil and the amount of compounds such as a-pinene, cineol and linalool were reported in the mature and young leaves' volatile oil.

Keywords: Eucalyptus species citriodora, Citronella, Biological control, Chromatographic Gas.

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INTRODUCTION:
The Myrtaceae family represents a rich source of essential oils with various biological activities including antibacterial, antifungal and anti-inflammatory effects. The Myrtaceae various species with potent antimicrobial effect and their volatile oils as the antimicrobial. Antifungal and agents are used in the creams, soaps and toothpastes [1,2]. In this family, the Eucalyptus genus has been cultivated and utilized for many years [3]. The leaves of more than 300 species contain volatile oils and less than 20 species of this genus are commercially utilized for oil production [4]. Among the vast collections of herbs containing secondary metabolites especially volatile oils, are considered as one of the most important sources of the aforementioned materials due to the wide area under cultivation, adaptation to different climates, low food needs and rapid growth. The Australian continent which is about 7700000 square kilometers, is the main home for the Eucalyptus (Jahanshir and Mesdagh 1351). The various types of eucalyptus have a significant economic importance in wood industry due to the hardness, severity, durability and quality of wood with its rapid growth [5] and its lack of wood decay [6], suitable fuel source and construction materials in the food industry such as honey production, it is used as a wind breaker in the farm or as an ornament in parks and decoration by roads side and streets in the different countries [5].

It is also important for having the essential oils secretion bags, the stem, leaf, flower and fruit in the pharmaceutical and perfume industries [6]. Eucalyptus is a native Australian plant that is found mostly in tropical and temperate regions of northeastern Australia as well as is elegant with a smooth shiny slightly sticky body and in summer it is more colored with copper and has a little crown and thorns with scented lemon leaves have a pale skin color from silver to purple. The tree is tolerant with rapid growth but in young age it does not tolerate frost. The tree is widely welcomed in different countries to decorate and cultivate on the street or windbreaker [5]. The growing tree has 35 meters height. In Brazil, it is utilized to extract essential oils. The crown of the tree is quite open and the leaves smell a lot of lemon. Pear-shaped buds are connected in form of 3 clustered faces in the bottom of the leaf to the stem and fruits are jars shaped, this species is known as Lemon-Scented Gum, Blue-Spotted Gum and Eucalyptus citriodora.

MATERIALS AND METHODS:
Identification of Essential Oils Ingredients
In this experiment, the volatile oil of young and mature eucalyptus leaves was extracted utilizing a Clevenger apparatus. For identification of essential oils, GC gas chromatography and gas chromatography coupled to GC-MS mass spectrometry was used. After injecting the essential oil into the above devices utilizing the inhibition time of the compounds, the Kavats KI inhibitory index and the mass spectrum as well as the comparison of these components with standard compounds or with the information in the library of the device, the identification of the compounds of Essential oil was performed.

Specifications of the utilized devices Clevenger
The Clevenger machine is utilized to extract volatile oil from the sample. The device consists of three parts a flask, an oil separatory tub and a condenser.

It is better to keep the plant covered with water inside the balloon and does not touch the balloon because the direct air of the balloon causes the plant to burn and create an unpleasant odor as well as damage to the actual percentage of organic compounds.

GC Gas Chromatography
The Shimadzu gas turbine generator (GC-9A) equipped with F.I.D detector (hydrogen flame ionization) and DB-1 column with a length of 30 m and internal diameter of 0.25 microns was utilized. The thermal design of the column increased from 5 ° C / min to 15 ° C / min and helium gas was utilized at 1.1 mg / min as carrier gas.

Gas chromatography connected to GC-MS mass spectrometer
Thermoquestfinnigan gas chromatography of the Trace model connected to a mass spectrometer equipped with a DB-1 column with a length of 30 m and an internal diameter of 0.25 mm and a thin layer of a stationary phase of 0.25 micrometers was used. The column’s thermal design was similar to the column layout in the GC device and helium gas was utilized at a speed of 1.1 milliliters per minute as the carrier gas. The scanning time was equal to one second and the ionization energy was 70 electron volts.

RESULTS AND DISCUSSION:
The yield of essential oil of young and adult Eucalyptus cytriodora leaves was 0.6 and 0.4% respectively and 11 compounds were identified. Therefore, the distribution of the composition of the essential oil of the eucalyptus oil of the target species in Table. 1, shows that the amounts of a-pinene, cineol and linalool in young and mature leaves were more or less identical indicating a greater stability of these materials against environmental factors over time. In other words,
they have less volatility or lack of conversion to other materials during leaf growth. While the amount of compounds such as beta-pinene, terpine-4-α, α-Terpineol citrnel and citronelic acid indicate that these compounds are elevated in mature leaves compared to young leaves, then the amount of para-cymene and citronella content is 78% and 49. 77% showing a higher amount of these two compounds in young leaves than the mature leaves. Concerning the compounds of Beta-Pinene and Citronellol, it can be stated that over time the vegetative growth of the eucalyptus leaves of the specified species is approximately twice as high in the adult leaf which indicates the conversion of other materials into these two substances. In 1999, Citronella was reported as 75% by the Moudachirou & Gbenous [7] as the main constituent of essential oil of the eucalyptus leaves of the citriodora. In addition, compounds such as Citronella 73.3% and Citronellol 10.9% were the main components of Eucalyptus citriodora mentioned by Dagne et al. in 2000[8]. The essential oil yield of Eucalyptus citriodora leaves was reported to be 0.6% in 2006 by Batish et.al[9]. and the main components of the essential oil of the leaves, citronella 52.2% citronellol 12.3% and isopoulglol 11.9% announced. Based on the research performed out by other researchers and the present study, the citronella and citronelollol can be considered as the most important constituents of Eucalyptus cytriadora, but the reason for the difference in the amount of these compounds depends on the geographical situation, weather conditions, accumulation season and the conditions of the decoction and phytochemical analysis of the essential oil. It is worth to mentioned that in this study after the two combinations mentioned in the adult leaf cineol is 7% of the major combination that the main combination of volatile oils of many Eucalyptus species has been reported by many researchers in many sources which as Primary raw material is utilized for the various pharmaceutical cosmetic and sanitary industries (Carmenetal. 2003)[10].

CONCLUSIONS AND SUGGESTIONS:
Since, there is a direct relation between the compounds in each essential oil and its antimicrobial characteristics, the amount of these compounds can be ascertained by the decomposition of the essential oils and the major component or index that plays the main role is diagnosed and in the various researches carried out in various fields of science, it is applied to the application of natural plant products.

**Table 1:** Chemical Composition and Their Values for Young and Mature Eucalyptus Leaves Oil of citriodora

<table>
<thead>
<tr>
<th>mature leaf</th>
<th>young leaf</th>
<th>composition</th>
<th>row</th>
</tr>
</thead>
<tbody>
<tr>
<td>1/59</td>
<td>1/20</td>
<td>α-pinene</td>
<td>1</td>
</tr>
<tr>
<td>1/69</td>
<td>0/61</td>
<td>β-pinene</td>
<td>2</td>
</tr>
<tr>
<td>7/00</td>
<td>7/13</td>
<td>Cineol</td>
<td>3</td>
</tr>
<tr>
<td>0/83</td>
<td>0/66</td>
<td>Linalool</td>
<td>4</td>
</tr>
<tr>
<td>65/23</td>
<td>77/49</td>
<td>Citronellal</td>
<td>5</td>
</tr>
<tr>
<td>1/10</td>
<td>0/27</td>
<td>Terpine-4-ol</td>
<td>6</td>
</tr>
<tr>
<td>4/73</td>
<td>2/63</td>
<td>α-Terpineol</td>
<td>7</td>
</tr>
<tr>
<td>9/87</td>
<td>5/11</td>
<td>Citronellol</td>
<td>8</td>
</tr>
<tr>
<td>3/26</td>
<td>2/66</td>
<td>Citronelic acid</td>
<td>9</td>
</tr>
<tr>
<td>0/00</td>
<td>0/78</td>
<td>ρ-cymene</td>
<td>10</td>
</tr>
<tr>
<td>4/70</td>
<td>1/46</td>
<td>Unidentified</td>
<td>11</td>
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REFERENCES: