INTRODUCTION

Essential oil is a concentrated liquid, contains volatile chemical compounds derived from medicinal plants, which often used as additives in the formulation of foods, beverages, perfumes, medicines, and cosmetics. One of the medical plants that have volatile essential oils is *Eucalyptus globulus* Labill., which belongs to the Myrtaceae family and is native to Australia and nowadays cultivate worldwide (Dhakad et al., 2018; Aziz et al., 2018). *E. globulus* is a tree that naturally grows to a height of 60-80 m. The bark of plant with brown or yellowish-brown color is mostly smooth, shedding in long strips to leave a white or greyish surface that is resistant to rot and can easily separate from the stem. Its leaf color also depends on the age of the tree (initially blue-green and over time greenish-white) (Pereira et al., 2014; Tyagi et al., 2011; Araujo et al., 2010). The flower buds are organized singly in leaf axils on a condensed peduncle that is sessile or up to 5 mm long. The peak of flowering occurs between May and January, which are white with numerous stamens in white or cream or yellow or red color.

Additionally, the fruits of *E. globulus* are a woody semicircular capsule form with the valves close by edge level (Mason et al., 2000; Mulyaningisih et al., 2010; Nakhaei et al., 2009). Eucalyptus's therapeutic properties include antiseptic, anti-parasitic, insect repellent, anti-rheumatism, anti-migraines, anti-urinary tract infections, anti-ulcer burns, febrifuge, and anti-fatigue (Adebola et al., 1999; Abdollahzade et al., 2012; Zaifton et al., 2018). Since eucalyptus leaves are widely used in this region, it is important to identify their...
chemical constituents. Eucalyptus essential oil widely uses as a traditional medicine in many parts of the world to the treatment of different diseases and disorders (Zaiton et al., 2018). Previous studies showed that prescription of traditional doses of E. globulus essential oil dissolved in water by transdermal rout up to 4–7 g/100 L had an impressive effect (Committee on Herbal Medicine Product, 2013).

The most prominent extraction compounds of the Eucalyptus leaf include 1, 8-Cineole, and Eucalyptol (Adebola et al., 1999; Abdollahzade et al., 2012). Phytochemical studies have shown that the main ingredient of the E. globulus essential oil is 1, 8-Synolol (Silva et al., 2003). Eucalyptol or 1, 8-Cineole is a compound with the chemical formula C_{10}H_{18}O is a compound with the identity of pub. Its molar mass is 154.249 g/mol. There are various other names for this combination of liquid and colorless natural organic such as Cajeputol, Oxido-p-menthane and Limonen-1, 8-oxid. 1, 8-Cineole has phlegmic, bactericidal and anti-inflammatory effects. It is a cyclic ether and a monoterpen. Eucalyptol is a natural constituent of a number of aromatic plants and their essential oil fraction (Silva et al., 2003). Whereas many varieties of medicinal plants are rich in odorant and volatile compounds, and these volatile oils have significant roles in the pharmaceutical, agrochemical, aromatherapy, and food flavorings of humans. The aim of the present study was investigation the chemical composition of the essential oil of E. globulus leaves collected from Dehloran in Ilam province, Iran as an important geographical zone for medicinal plants, by Head space Solid Phase Microextraction (HS-SPME) method.

MATERIALS AND METHODS

Preparation of medicinal plants
Eucalyptus leaves (Figure 1) collected from the Dehloran district (a latitude of 32°41'31.91"N and a longitude of 47°16'4.64"E or 32.692197 and 47.267956 respectively), Ilam province located in western of Iran during the October 2019. The Eucalyptus leaves dried for 72 hours at a standard temperature. Then, using a mixer powder prepared and finally, the essential oils and its volatility examined and characterized by the solid phase microextraction (HS-SPME) method.

Identification of chemical compounds by HS-SPME
In this experiment, the essential oil of the plant was extracted by HS-SPME technique. In this technique, about 2 grams of dried plant and its powder were placed in the vial and the vial temperature was set at 60–70 °C. These optimum temperature conditions will saturate the vapor content of the substances in the plant essential oil in the headspace of the solid surface. The SPME syringe with a lid on it was then placed in the headspace of the container and the material in the vapor was absorbed by the silica phase in the instrument needle. After the silica fiber was allowed to sufficiently saturate with volatile components, the fiber was directly placed into the GC/MS input section and materials present in the fiber were adsorbed due to the temperature of the input and then entered into the GC/MS apparatus for identification (Hashemi et al., 2012).

HS-SPME METHOD
2 g of each plant extract was used for analysis. The device condition was as follows: Gas chromatograph (Agilent6890N) was coupled to Agilent 5973 Mass detector; Column: HP - 5. (30 m length × 0.25 mm (ID) × 0.25 μm (stationary phase thickness); Injector type: split/ splitless and column temperature program: 50 °C, hold time 0.00 min and rate of -5°C/min; temperature 200 °C, hold time, 0.00 min and rate of 5°C/min and temperature 240 °C, hold time 0.00 min and rate of 10 °C/min. Carrier gas: He (99.999%); Injection type: splitless; Library: Wiley 7n;
Table 1: Identified compounds of Eucalyptus essential oil by HS-SPME (GC-MS).

| Retention time | Compound                  | Molecular formula | Chemical structure | %    |
|----------------|---------------------------|-------------------|--------------------|------|
| 5.817          | Tricyclene                | C_{10}H_{16}       | ![Tricyclene](image) | 0.24 |
| 6.103          | α-Pinene                  | C_{10}H_{16}       | ![α-Pinene](image)  | 0.15 |
| 6.463          | Camphene                  | C_{10}H_{16}       | ![Camphene](image)  | 3.77 |
| 6.6            | Verbenene                 | C_{10}H_{14}       | ![Verbenene](image) | 1.67 |
| 7.074          | Sabinene                  | C_{10}H_{16}       | ![Sabinene](image)  | 0.06 |
| 7.16           | β-pinene                  | C_{10}H_{16}       | ![β-pinene](image)  | 0.57 |
| 7.406          | 1-Octen-3-ol              | C_{8}H_{16}O       | ![1-Octen-3-ol](image) | 1.22 |
| 7.503          | β-Myrcene                 | C_{10}H_{16}       | ![β-Myrcene](image) | 0.55 |
| 7.886          | 1-Phellandrene            | C_{10}H_{16}       | ![1-Phellandrene](image) | 0.06 |
| 8.017          | 3-Carene                  | C_{10}H_{16}       | ![3-Carene](image)  | 0.40 |
| 8.212          | α-Terpinene               | C_{10}H_{16}       | ![α-Terpinene](image) | 0.07 |
| 8.446          | P-Cymene                  | C_{10}H_{16} or C_{10}H_{16} | ![P-Cymene](image) | 1.04 |
| 8.772          | 1,8-Cineole               | C_{10}H_{14}O      | ![1,8-Cineole](image) | 51.25 |
| 8.749          | cis-Ocimene               | C_{10}H_{16}       | ![cis-Ocimene](image) | 1.85 |
| 9.035          | trans-β-Ocimene           | C_{10}H_{16}       | ![trans-β-Ocimene](image) | 0.36 |
| 9.36           | α-Terpinene               | C_{10}H_{16}       | ![α-Terpinene](image) | 0.15 |
| 9.72           | trans-Sabinene hydrate    | C_{10}H_{10}       | ![trans-Sabinene hydrate](image) | 0.39 |
| 10.166         | Terpinolen                | C_{10}H_{16}       | ![Terpinolen](image) | 0.32 |
| 10.641         | Linalool                  | C_{10}H_{18}O or C_{10}H_{18} | ![Linalool](image) | 1.79 |
| 11.944         | Camphor                   | C_{10}H_{20}O      | ![Camphor](image)   | 9.58 |
| 12.069         | Isomenthone               | C_{10}H_{18}O      | ![Isomenthone](image) | 1.51 |
| 12.327         | MEN-THOFURAN              | C_{10}H_{18}O      | ![MEN-THOFURAN](image) | 1.30 |
| 12.715         | Bornol                    | C_{10}H_{18}O      | ![Bornol](image)    | 7.63 |
| 12.881         | 4-Terpineol               | C_{10}H_{18}O      | ![4-Terpineol](image) | 0.25 |
| 13.15          | Krypton                   | Kr                 | 1587485            | 0.35 |
| 13.738         | Eucarvone                 | C_{10}H_{18}O      | ![Eucarvone](image) | 0.29 |
| 14.15          | Isobornyl acetate         | C_{12}H_{20}O      | ![Isobornyl acetate](image) | 1.01 |
| 14.584         | Cuminic aldehyde          | C_{15}H_{12}O      | ![Cuminic aldehyde](image) | 0.17 |
| 14.67          | Carvone                   | C_{10}H_{14}O      | ![Carvone](image)   | 0.17 |
| 14.83          | Linalyl acetate           | C_{12}H_{20}O or C_{12}H_{20}O | ![Linalyl acetate](image) | 1.91 |
| 15.699         | Bornyl acetate            | C_{12}H_{20}O      | ![Bornyl acetate](image) | 1.49 |
| 15.887         | Menthyl acetate           | C_{15}H_{22}O      | ![Menthyl acetate](image) | 1.86 |
| 18.528         | β-elemene                 | C_{15}H_{26}       | ![β-elemene](image) | 0.41 |
| 18.956         | α-Gurjunene               | C_{15}H_{24}       | ![α-Gurjunene](image) | 0.10 |
| 19.248         | trans-Caryophyllene       | C_{15}H_{24}       | ![trans-Caryophyllene](image) | 1.33 |
| 19.928         | δ-Cadinene                | C_{15}H_{24}       | ![δ-Cadinene](image) | 0.09 |
| 20.071         | Trans-beta-Farnesene      | C_{15}H_{24}       | ![Trans-beta-Farnesene](image) | 1.15 |
| 20.808         | Germacrene D              | C_{15}H_{24}       | ![Germacrene D](image) | 0.82 |
| 20.911         | β-ION-ONE                 | C_{15}H_{26}O      | ![β-ION-ONE](image) | 0.11 |
| 21.368         | Geranyl acetate           | C_{15}H_{26}O      | ![Geranyl acetate](image) | 0.34 |
| 25.158         | alpha-Amorphene           | C_{15}H_{24}       | ![alpha-Amorphene](image) | 1.10 |
| 23.357         | Caryophyllene oxide       | C_{15}H_{24}       | ![Caryophyllene oxide](image) | 0.66 |
| 24.74          | α-Cadinol                 | C_{15}H_{26}O      | ![α-Cadinol](image) | 0.48 |
Injector temperature: 250 °C and flow rate: 0.9 mL/min. Extraction mode: (HSSPME); SMPE fiber: PDMS 100 μm thickness (SUPELCO); sample weight: 0.5 g; extraction temperature: 60 °C; extraction time: 20 min; sonication time: 10 min (Euronda sonication instrument, Italy) and desorption time in GC-MS injector port: 3 min (Bahmani et al., 2019).

RESULTS

In the beginning, the volatile essential oils extracted by solid-phase microextraction in the head space (HS-SPME), and then the essential oils analyzed by GC-MS. Based on the results, 43 compounds identified by the HS-SPME method in the isolated essential oil. 1, 8-cineole with 51.25% was the highest chemical composition of Eucalyptus essential oil (Table 1, Figure 1). The other important identified compounds of Eucalyptus essential oil were camphor (9.58%), borneol (7.63%), and camphene (3.77%), respectively. Other information and details of the identified chemical compounds mentioned in Table 1 and Figure 1.

Properties of 1, 8-Cineole such as chemical formula, Molar mass, Density, Melting point, Boiling point and Magnetic susceptibility (χ) listed in Table 2.

| Properties of 1, 8-Cineole | C_{10}H_{18}O |
|---------------------------|---------------|
| Molar mass                | 154.249 g/mol |
| Density                   | 0.9225 g/cm³  |
| Melting point             | 2.9 °C (37.2 °F; 276.0 K) |
| Boiling point             | 176–177 °C (349–351 °F; 449–450 K) |
| Magnetic susceptibility (χ)| −116.3×10⁻⁴ cm³/mol |

The results achieved from the chromatogram in Figure 1 revealed that the essential oil of Eucalyptus comprises 43 compounds. The major components identified include bicyclic monoterpenes, aromatic terpenoids, cyclic ether, and terpenes.

DISCUSSION

Until now, over 700 tree species and bushes belonging to the Eucalyptus recognize in the world. It belongs to the family Myrtaceae and found in most tropical regions of the world (Boland et al., 1991). In this study, Eucalyptus leaf collected from Dehloran city, the western region of Iran, located in the Ilam province. Based on obtained results and in agreement with previous studies, the major ingredient of volatile essential oils of Eucalyptus species is 1, 8-cineole (Carmen et al., 2003).

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AUTHORS CONTRIBUTION

MB reviewed the literature and prepared the first draft of manuscript; ZKH, ZE, MB and NA reviewed the
CONFLICT OF INTERESTS

All authors have declared no conflict of interest.

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