Comparison of Identified Compounds from Extracted *Pelargonium Radula* Leaves by Supercritical Fluid Extraction and Commercial Geranium Essential Oil

I F Gaaffar¹, N A M Zainuddin¹, and S Zainal¹

¹ Faculty of Chemical Engineering, Universiti Teknologi MARA (UiTM), Shah Alam, Selangor, Malaysia.

Corresponding author e-mail: nurain1465@uitm.edu.my

Abstract. Supercritical fluid extraction (SFE) is a green technology that is convenient to extract *Pelargonium radula* (*P. radula*) leaves without leaving any negative impacts on the environment. It is an alternative approach to reduce the solvent residual problem during the extraction process. Therefore, this research was performed to compare the active ingredients extracted by SFE technique with the commercialized geranium essential oil in the market. Extraction of powdered *P. radula* leaves by supercritical carbon dioxide (SC-CO₂) was operated at constant pressure, temperature, and solvent flowrate which were 100 bar, 40 °C, and 24 mL/min, respectively. SC-CO₂ extraction on *P. radula* leaves result in 0.19 % of oil yield. By comparing extracted *P. radula* oil with the commercialized geranium essential oil, both oil consists of benzyl acetate, citronellol, geraniol, e-amyl cinnamaldehyde, isopropyl tetradecanoate, and isopropyl hexadecanoate.

1. Introduction

Essential oils are extracts from certain segment of plants that contains oil. The use of essential oils has gained tremendous attention among the community at every age. Essential oils have been applied in various industries nowadays such as in perfumery, food, pharmaceutical, and aromatic industries. *P. radula* is one of *Pelargonium sp.* plant that is belongs to the *Geraniaceae* family and cultivated from South Africa [1,2]. Based on the physical appearance of *P. radula*, it has a height between 0.75 m to 1 m and hairy texture can be felt on the surface of the leaves that emits pleasant odour similar to rose and lemon [3,4].

Apart from producing oxygen for humans and animals to breathe, maintaining air balance and supplying food resources, plants also might influence people’s health and emotions. *P. radula* plant has its own specialty. Traditionally, diseases such as tonsillitis, diabetes, malaria, abdominal, and uterine disorders are being treated either by consuming the raw *Pelargonium* leaves or boiled to drink [4,5]. Besides, geranium oil that is obtained from the *Pelargonium* leaves also helps people who have difficulty in sleeping, high blood pressure, low metabolism, as well as emotional and behavioural disorder [2].

Recently, conventional extraction methods either hydrodistillation, solvent extraction, or steam distillation has always been the main choice for the researchers to extract bioactive components from aromatic and medicinal plants because of their simplicity. Nonetheless, there are several weaknesses...
associated with these conventional extraction techniques, and one of them is time-consuming. There are a great numbers of studies showing that conventional extraction methods take quite a long time to complete. For instance, Jalali-Heravi et al. [6] implemented hydrodistillation technique that took 360 minutes to extract Iranian geranium plant. Meanwhile, Asnawi et al. [7] used solvent extraction and steam distillation method to extract P. radula oil which took 2880 minutes and 600 minutes, respectively. Furthermore, practicing conventional extraction methods leaf a large amount of solvent residual at the end of extraction process. Thus, an additional process is required to be done after extraction process has been completed, which is through the separation of solvent from the extract [8,9]. These proved that conventional extraction methods have shortcomings in terms of extraction time that take too long to be completed. In addition, conventional extraction methods are well-known for its non-selective and non-environmentally friendly process. Based on research conducted by Viganó et al. [10], Soxhlet extraction method is considered as less selective technique as the concentration of antioxidant extracted from passion fruit bagasse is low. Plus, the usage of organic solvent especially in solvent extraction method is hazardous, flammable and toxic [11]. There are numerous studies that have been performed that used organic solvent such as methanol and ethanol to extract valuable components. For instance, Bimakr et al. [12] uses pure ethanol, methanol, petroleum ether and 70 % ethanol to extract major bioactive flavonoid compounds from Mentha spicata L. leaves. However, Budisa & Schulze-Makuch [13] have agreed that organic solvent such as alcohol might cause an adverse impact on human’s health and environment, extremely.

SFE is a type of extraction technique that uses solvent in supercritical state to extract the beneficial compounds found in medicinal and aromatic plants. Currently, SFE gains much attention from the researchers due to its effectiveness and environmental friendly process. Frequently, CO$_2$ is considered to be used as solvent for SFE method as CO$_2$ has low critical conditions in which the critical temperature and pressure for CO$_2$ are 31.1 $^\circ$C and 73.8 bar, respectively [14]. The existence of SFE technology were able to overcome the weaknesses of conventional extraction techniques especially in terms of extraction time and operating temperature. Based on the recent research, there have been numerous studies have proven that SFE offers shorter extraction time compared to conventional extraction technique. As evidences, Idris et al. [15] took only 45 minutes to complete the extraction of tamarind seed while Zainuddin et al. [16] took 60 minutes to extract Leucaena leucocephala pod oil. This proved that SFE method is better in terms of time as compared with conventional extraction methods. In the context of selectivity, recent studies have proved that SFE is more selective compared with the conventional extraction methods as this technique is capable of extracting high concentration of volatile compounds [17]. Furthermore, CO$_2$ is considerable to be used as solvent as it has the characteristics of non-flammable, non-toxic, selective, odourless, and safe to be used [18]. Therefore, this research was performed to study the capability of SFE method to extract P. radula oil and compare the extracted components in the oil with the commercialized geranium essential oil in the market.

2. Materials and method

2.1. Equipment and chemicals

SFE extraction process was conducted using Supercritical Fluid Technologies Model SFT-100 equipment. SFE equipment consists of a high-pressure delivery pump, a preheat coil, 25 mL of extraction vessel, a static and dynamic valve, and a restrictor valve as shown in Figure 1. Schematic diagram of SFT-100. The CO$_2$ tank of SFC grade with 99.99% purity was purchased from SR Focus Trading. Meanwhile, chromatographic analysis of P. radula oil was performed using Agilent Technologies 7890A/5975C Series MSD apparatus that equipped with a non-polar Agilent HP-5MS column with a length of 30 m, 0.25 mm diameter and 0.25 $\mu$m thickness.
2.2. Sample preparation

*P. radula* plants were purchased from a nursery in Sungai Buloh. The leaves, which is the main interest of the study is first separated and washed with distilled water to remove any impurities. The leaves were cut into small pieces and then dried in a Memmert UFE 500 oven at 45 °C for 24 hours until the moisture content of the sample below 10 %. The dried sample was then ground by using mechanical grinder model MD30-CS. Meanwhile, geranium essential oil was bought from Best Formula Industries.

2.3. Supercritical fluid extraction

SFE process was conducted using a laboratory-scale equipment and CO\textsubscript{2} was used as a solvent to extract *P. radula* oil. An amount of 6 g powder *P. radula* leaves was inserted in a cotton sachet and loaded into the extraction vessel. The extraction process was performed at a constant pressure, temperature, and solvent flowrate which were 100 bar, 40 °C, and 24 mL/min, respectively, throughout the experiment. The extraction process ran for 70 minutes and the oil collection was done for every 10 minutes throughout the period of the extraction process. Once the oil has been collected in a glass vial, the mass of oil extracted was determined and the percentage of yield was calculated by using equation (1).

\[
\text{Yield (\%)} = \frac{\text{mass of extracted oil}}{\text{mass of sample}} \times 100
\]  

(1)

2.4. Gas chromatography-mass spectrometry (GC-MS) analysis of oil

A solution was prepared in which 1 mg of extracted *P. radula* oil was dissolved in 0.5 mL of hexane solvent. The organic fraction of extracted *P. radula* oil and commercialized *P. radula* essential oil were analyzed by GC-MS. The analytical condition with sample injection (1µL), carrier gas helium (99.999% pure) and flow rate 1 mL/min with a split mode on, with the ratio of 50:1 injector temperature 250 °C.
with the split flow of 50 mL/min. The oven temperature was set at 60 °C for 10 minutes, later increased at a rate of 3 °C /min to 230 °C which then needed to be maintained for 1 minute. The compounds detected were based on the mass spectra library of the National Institute of Standard Technology (NIST).

3. Results and discussion
SC-CO\textsubscript{2} extraction was conducted at fixed operating conditions which were 100 bar, 40 °C and 24 mL/min for 70 minutes. Every 10 minutes in the interval of extraction process, the extract of \textit{P. radula} was collected in a glass vial. Based on the mass of extracted oil, the percentage of extracted \textit{P. radula} oil was calculated by using equation (1). The extraction of \textit{P. radula} leaves by using SC-CO\textsubscript{2} was conducted twice to obtain an average result. Figure 2 shows the average percentage of extracted \textit{P. radula} leaves oil yield as a function of time. In terms of extraction time, it can be observed that at 60 minutes of extraction time, the extracted oil yield starts to remain constant. This shows that the extraction of \textit{P. radula} leaves only took 60 minutes to be completed in which the result obtained is in agreement with Zainuddin et al. [16]. Based on the outcome of the experiment, SC-CO\textsubscript{2} extraction was capable to extract 0.19 % of \textit{P. radula} oil yield. This result is quite similar to a study conducted by Gomes et al. [19] in which 0.22 % of rose geranium oil was yielded by using SFE process at 100 bar and 40 °C. It was found that oil yield in SFE is lower compared to conventional extraction method. Asnawi et al. [7] conducted conventional extraction methods on \textit{P. radula} leaves by using solvent extraction and steam distillation process. Both solvent extraction and steam distillation technique 7.94 % and 2.54 %, respectively [7]. This phenomenon may be because of low operating pressure and temperature of SC-CO\textsubscript{2} extraction process that result in low density of SC-CO\textsubscript{2}. The lower the density of SC-CO\textsubscript{2}, the lower the solubility of \textit{P. radula} oil to be extracted by SC-CO\textsubscript{2}. Nevertheless, another advantage of SFE technique is the solubility of solvent can be enhanced by varying the operating conditions such as pressure, temperature and solvent flow rate to maximise the oil extraction yield.

\begin{figure}[h]
\centering
\includegraphics[width=\textwidth]{figure2.png}
\caption{Yield of \textit{P. radula} extract at pressure of 100 bar, temperature 40 °C, and solvent flow rate of 24 mL/min for 70 minutes}
\end{figure}
Once the extraction process completed, the extracted *P. radula* oil and the commercialized geranium essential oil that has been purchased were analysed by GC-MS. Figure 3 and Figure 4 reveal the chromatogram for commercialized geranium essential oil and extracted *P. radula* oil. Meanwhile, Table 1 lists the compounds characterised by GC-MS. Based on the GC-MS analysis, six out of nine compounds were found to be similar between extracted *P. radula* oil and commercialized geranium essential oil. The six similar compounds were benzyl acetate, citronellol, geraniol, e-amyl cinnamaldehyde, isopropyl tetradecanoate, and isopropyl hexadecanoate. However, compounds like linalool, phenyl ethyl alcohol and e-anethole were not detected in the extracted *P. radula* oil by SFE technique due to selective characteristics of SC-CO$_2$ [11,20,21]. This result is compatible with the findings by Ghasemi et al. [22] which conclude that SFE method has better selectivity especially to reduce the extraction of unwanted compounds. Additionally, linalool is formed from the degradation of the geraniol compound [19]. As an evidence for the absence of linalool compound in the extracted *P. radula* oil might due to extraction process was performed at low temperature which was at 40 °C, thus, the degradation of geraniol into linalool does not occur.

**Figure 3.** Chromatogram of commercialized geranium essential oil

**Figure 4.** Chromatogram of extracted *P. radula* oil by SC-CO$_2$ at 100 bar, 40 °C, and 24 mL/min
| Peak | Retention time (min) | Name of compound             | Area of peak (%) | Commercialized geranium essential oil | Extracted $P. radula$ oil by SFE |
|------|----------------------|------------------------------|------------------|---------------------------------------|----------------------------------|
| 1    | 18.204               | Linalool                     | 0.79             | -                                     |                                  |
| 2    | 19.114               | Phenyl ethyl alcohol         | 3.75             | -                                     |                                  |
| 3    | 21.941               | Benzyl acetate              | 0.41             | Trace                                 |                                  |
| 4    | 23.829               | $E$-anethole                 | 0.71             | -                                     |                                  |
| 5    | 25.740               | Citronellol                  | 2.64             | Trace                                 |                                  |
| 6    | 27.062               | Geraniol                    | 1.23             | Trace                                 |                                  |
| 7    | 44.565               | $E$-amyl cinnamaldehyde     | 0.27             | Trace                                 |                                  |
| 8    | 51.518               | Isopropyl teradecanoate     | 66.84            | Trace                                 |                                  |
| 9    | 58.518               | Isopropyl hexadecanoate     | 15.14            | 25.73                                 |                                  |

4. Conclusion
This research was conducted to compare the extracted components from $P. radula$ leaves by SFE and the geranium essential oil that have been commercialized in the market. $P. radula$ leaves were extracted at constant pressure, temperature and solvent flowrate, which were 100 bar, 40 °C, and 24 mL/min, respectively, for 60 minutes of extraction time. Next, the extracted $P. radula$ oil and the commercialized geranium essential oil were analysed by using GC-MS. Based on the experiment, 0.19 % of oil yield was successfully being extracted by SFE method. In terms of extracted components, both of the oils consist of 6 similar compounds which are benzyl acetate, citronellol, geraniol, $E$-amyl cinnamaldehyde, isopropyl tetradecanoate, and isopropyl hexadecanoate. However, the selectivity properties of $CO_2$ as a solvent for supercritical fluid extraction technique is an evidence to show that the number of components extracted $P. radula$ oil is less than the commercialized geranium essential oil.

5. Acknowledgment
The authors are grateful and thanked every individual that is involved in this project especially lab technician of Faculty of Chemical Engineering UiTM Shah Alam and Integrated Separation Technology Research Group (i-STRONG) for their technical support. A massive thanks to Bestari UiTM for the financial assistance provided through a research grant that is Bestari UiTM (Code: 600IRMI/DANA 5/3/BESTARI (126/2018)).

Reference
[1] Kassahun B M, Zigene Z D and Teferi Z 2012 Yield and yield components of rose scented Geranium (Pelargonium graveolens) as influenced by plant population density in Ethiopia 2 60–8
[2] Jaggali S, Venkatesh K, Babu Rao N, Hilal M H and Rani A R 2011 Phytopharmacological importance of Pelargonium species J. Med. Plants Res. 5 2587–98
[3] Peterson A, Machmudah S, Roy B C, Goto M, Sasaki M and Hirose T 2006 Extraction of essential oil from geranium (Pelargonium graveolens) with supercritical carbon dioxide J. Chem. Technol. Biotechnol. 81 167–72
[4] Sharopov F S, Zhang H and Setzer W N 2014 Composition of geranium (Pelargonium graveolens) essential oil from Tajikistan Am. J. Essent. Oils Nat. Prod. AJEONP 2 13–6
[5] Asgarpanah J and Ramezanloo F 2015 An overview on phytopharmacology of Pelargonium graveolens L Indian J. Tradit. Knowl. 14 558–63
[6] Jalali-Heravi M, Zekavat B and Seresht H 2006 Characterization of essential oil components of Iranian geranium oil using gas chromatography-mass spectrometry combined with chemometric resolution techniques. J. Chromatogr. A 1114 154–63

[7] Asnawi S, Mohd Zaki Z, Abdul Azizi A, Khamis A K and Abdul Aziz B 2008 Evaluation of the potential of Pelargonium Radula extract in repelling Aedes aegypti. J. Chem. Nat. Resour. Eng. 11–9

[8] Guedes A R, de Souza A R C, Zanoelo E F and Corazza M L 2018 Extraction of citronella grass solutes with supercritical CO2, compressed propane and ethanol as cosolvent: Kinetics modeling and total phenolic assessment. J. Supercrit. Fluids 137 16–22

[9] Montañés F, Catchpole O J, Tallon S, Mitchell K A, Scott D and Webby R F 2018 Extraction of apple seed oil by supercritical carbon dioxide at pressures up to 1300 bar. J. Supercrit. Fluids 141 128–36

[10] Viganó J, Coutinho J P, Souza D S, Baroni N A F, Godoy H T, Macedo J A and Martínez J 2016 Exploring the selectivity of supercritical CO2 to obtain nonpolar fractions of passion fruit bagasse extracts. J. Supercrit. Fluids 110 1–10

[11] Sahena F, Zaidul I S M, Jinap S, Karim A A, Abbas K A, Norulaini N A N and Omar A K M 2009 Application of supercritical CO2 in lipid extraction - A review. J. Food Eng. 95 240–53

[12] Bimakr M, Rahman R A, Taip F S, Ganjloo A, Salleh L M, Selamat J, Hamid A and Zaidul I S M 2011 Comparison of different extraction methods for the extraction of major bioactive flavonoid compounds from spearman (Mentha spicata L.) leaves. Food Bioprod. Process. 89 67–72

[13] Budisa N and Schulze-Makuch D 2014 Supercritical Carbon Dioxide and Its Potential as a Life-Sustaining Solvent in a Planetary Environment Life 4 331–40

[14] Yunus M A C, Arsad N H, Zhari S, Idham Z, Setapar S H and Mustaph A N 2013 Effect of supercritical carbon dioxide condition on oil yield and solubility of Pinus palustris seeds. J. Teknol. (Sciences Eng.) 60 45–50

[15] Idris S A, Jantan S, Alias N H, Yusof N M, Ghazali N A and Yahya E 2015 Solubility Determination of Tamarind Seeds Extracts by Using Supercritical CO2 Extraction

[16] Zainuddin N A, Subuki I, Ibrahim K A, Ismail N F, Yatim S R M and Razak N H A 2018 Supercritical carbon dioxide extraction of Leucaena leucocephala Pod-Oil Yield & component identification. Int. J. Eng. Technol. 7 65–70

[17] Machalova Z, Sajfrtova M, Pavela R and Topiar M 2015 Extraction of botanical pesticides from Pelargonium graveolens using supercritical carbon dioxide. Ind. Crops Prod. 67 310–7

[18] Sapkale, G.N. & Patil, S.M. & Surwase, U.S. & Bhatbhage P . 2010 Supercritical fluid extraction - a review Int. J. Chem. Sci. 8 729–43

[19] Gomes P B, Mata V G and Rodrigues A E 2007 Production of rose geranium oil using supercritical fluid extraction. J. Supercrit. Fluids 41 50–60

[20] Zeković Z, Bera O, Đurović S and Pavlić B 2017 Supercritical fluid extraction of coriander seeds: Kinetics modelling and ANN optimization. J. Supercrit. Fluids 125 88–95

[21] Khajeh M 2011 Optimization of process variables for essential oil components from Satureja hortensis by supercritical fluid extraction using Box-Behnken experimental design. J. Supercrit. Fluids 55 944–8

[22] Ghasemi E, Raofie F and Najafi N M 2011 Application of response surface methodology and central composite design for the optimisation of supercritical fluid extraction of essential oils from Myrtus communis L. leaves. Food Chem. 126 1449–53