Physicochemical Parameters and Chemical Compositions of Essential Oils from Marjoram and Sweet Basil Cultivated in Northern Thailand

Nanthaphong Khamthong*, Krisana Pootakham, and Natwarin Krittiyapichartkul

College of Oriental Medicine, Rangsit University, Pathum Thani 12000, Thailand
*Corresponding author, E-mail: nanthaphong.k@rsu.ac.th

Abstract

Several aromatic plants have been experimentally cultivated and promoted in northern Thailand at the Royal Agricultural Stations; two of them are marjoram and sweet basil. Apart from supplying their fresh leaves for foods, they are also for essential oil production. However, there has been no report on the quality of the essential oils from these two plants cultivated in Thailand, which prompted us to investigate the quality of essential oils from marjoram and sweet basil cultivated in the highland area of Thailand. The overground parts of marjoram and sweet basil were collected from the Royal Agricultural Stations in Chiang Mai Province and subjected to hydrodistillation. The physicochemical parameters and chemical compositions of the hydrodistilled oils were determined and compared with those of the commercial ones. TLC and GC-MS were employed for the chemical analysis of the essential oils. The results demonstrated that their physicochemical properties (color, odor, specific gravity, refractive index, and solubility in ethanol) were not much different from the commercial ones. The GC-MS analysis revealed that the hydrodistilled marjoram oil contained 3-cyclohexen-1-ol (39.21%) as the principal component followed by γ-terpinene (9.00%) and *trans*-sabinene hydrate (1*R*-isomer) (9.00%). Of the commercial oil, however, 1,8-cineole (57.16%) and linalool (17.88%) were major compounds. Linalool (60.03%) and eugenol (19.62%) were mainly present in the hydrodistilled sweet basil oil, whereas estragole accounting for 85.79% was dominant in the commercial oil. This study exhibited that both the hydrodistilled marjoram and sweet basil oils were different from the commercial ones in terms of chemical compositions. Consequently, factors leading to the variation in the amount of the chemical components in the marjoram and sweet basil oils are worth further investigating. The essential oil from sweet basil cultivated at the Royal Agricultural Station could serve as a rich source of linalool.

Keywords**:** *physicochemical characteristics, chemical compositions, essential oils, marjoram, sweet basil*

1. Introduction

Plants containing essential oils and their products play important roles in daily life as medicines, cosmetics, aromatherapy and personal products, and the food industry. Most essential oils are very important ingredients in aromatherapy products that heal both mind and body in alternative medicines. Essential oils play different pharmacological activities, so they act as natural medicine (Ali et al., 2015). Marjoram and sweet basil are most commonly used fresh in foods, while their essential oils were employed in medicine and aromatherapy.

Marjoram or sweet marjoram (*Origanum majorana* L.) is a well-studied medicinal plant, commonly used as spices in foods. Marjoram and its oil have many pharmacological effects mainly associated with antimicrobial, anti-inflammatory, antioxidant, and relief of muscle cramps. Marjoram has been used as essential oil and extracts in food, medicine, aromatherapy, and personal health care products for their pharmacological activities (Bina & Rahimi, 2017). Similarly, sweet basil (*Ocimum basilicum* L.) is also well-investigated and used as spices in foods. Its pharmacological activities involved central nervous system (CNS) depressants beneficial for the alleviation of mental fatigue, colds, spasm, rhinitis, nausea, flatulence, and dysentery (Ch, Naz, Sharif, Akram, & Saeed, 2015).

Regarding differences in localities, plantation environment, climatic conditions, and so on, several studies revealed that these affected chemical components of the essential oils. For example, sampling of marjoram's aerial parts at different seasons in Egypt for essential oil distillation resulted in the variation in

the main components of the oil (Soliman, Yousif, Zaghloul & Okba, 2009). The major compounds detected in the oils in spring, summer, autumn, and winter were thymol, terpinene-4-ol, terpinolene, and *cis*-sabinene hydrate, respectively. A comparison of major components in the essential oils of marjoram from three different localities (Egypt, France, and Hungary) exhibited that the ratio between terpinene-4-ol to linalool and linalyl acetate increased in the order of Egyptian, French, and Hungarian marjoram (Krishnakumar & Potty, 2012). Baâtour et al. (2012) displayed that different culture conditions led to the bioconversion between *cis*- and *trans*-isomers of sabinene hydrate, a major component of the oil from the shoot. The one grown in a hydroponic medium contained *cis*-sabinene hydrate and terpinene-4-ol as major compounds. However, the other one was planted in inert sand consisted of *trans*-sabinene hydrate and terpinene-4-ol. Additionally, the same results were observed under saline treatments. Gharib, Moussa, and Massoud (2008) showed that the major compositions of Egyptian marjoram oil were *cis*-sabinene hydrate (18.47%) and terpinene-4-ol (24.24%), of which the relative percentages changed due to the fertilization type or level. Based on major components, it was suggested earlier that the oils of marjoram occurred in two types: one with terpinen-4-ol and sabinene hydrate and the other one with thymol and/or carvacrol (Vera & Chane-Ming, 1999).

Like the marjoram essential oils, the compositions of sweet basil oils can also vary depending on various parameters. For example, the oil from sweet basil cultivated in East Turkey having cold climate conditions had linalool as a major component (Agkul, 1989), whereas that grown in the tropical monsoon environment of Ethiopia predominated estragole (methyl chavicol) (Gebrehiwot, Bachetti, & Dekebo, 2016). Based on chemical composition and geographical origin, sweet basil oils have been classified into the following four groups: European basil oil (25.0-75.0% methyl chavicol and 25.4-50.0% linalool), Reunion basil oil (80% methyl chavicol), Tropical basil oil (>65% methyl cinnamate), and basil oil with a high percentage of eugenol (Akbari, Soltani, Binesh, & Amini, 2018).

In Thailand, certain amounts of dried aromatic plants of the temperate region together with their oils and extracts have been imported as ingredients in foods, medicine, aromatherapy, and personal care products. The Royal Agricultural Stations in northern Thailand have experimentally cultivated these plants intending to improve the quality of life of hill-tribe people and to substitute for some of those imported plants (Thailand Sustainable Development Foundation, 2016). Several aromatic plants are experimentally cultivated and promoted in northern Thailand to supply fresh leaves for foods and leftover parts for oils and extracts. Two of them are marjoram and sweet basil. Nevertheless, the good cultivation and harvesting yield of those two aromatic plants do not always contribute to the good quality of their essential oils, which relies on various factors such as plantation environment, climatic condition, and storage condition. There has been no report on the quality of the essential oils from marjoram and sweet basil cultivated in Thailand. Thus, the quality of these oils including physicochemical properties and chemical compositions must be determined and compared with the commercial ones. If the quality of the two essential oils obtained from the cultivation is comparable to that of the imported ones, they could be used to partly substitute those commercial oils in medicine, cosmetics, and aromatherapy products. Furthermore, the cultivation of these aromatic plants should be promoted among other crops in the highland area.

2. Objectives

The research aimed to investigate the physicochemical properties and chemical compositions of the essential oils from marjoram and sweet basil cultivated in northern Thailand and to compare the obtained data with those of the commercial oils.

3. Materials and Methods

3.1 Chemicals and reagents

Commercial marjoram and sweet basil oils were purchased from Thai-China Flavours and Fragrances Industry (TCFF) Co., Ltd. Analytical reagents including ethanol, ethyl acetate, and toluene were obtained from RCI Labscan, Thailand. HPLC grade ethanol, sulfuric acid, and vanillin were purchased from Merck, Germany. For TLC analysis, standard compounds including 1,8-cineole, terpinen-4-ol, geraniol,

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geraniol acetate, citral, and carvacrol were bought from Aldrich, USA, while myrcene, linalool, linalyl acetate, and camphor were provided from Fluka AG, Germany.

3.2 Plant materials

Marjoram (*Origanum majorana* L.) and sweet basil (*Ocimum basilicum* L.), of which the seeds were originally provided by Johnny's Selected Seeds (Maine, USA), were cultivated at the Royal Agricultural Stations at Pang Da, Samoeng distric,t and Khun Pae, Jomtong district, respectively, in Chiang Mai Province, Thailand. The overground parts were carefully collected and cleaned before further use.

3.3 Essential oil distillation

The hydrodistillation of the essential oils from the overground parts of marjoram (13.62 kg) and sweet basil (14.43 kg) was carried out using a Clevenger-type apparatus. The fresh plant materials were placed in a distillation flask and subjected to hydrodistillation for 2 hours. The oil was collected and separated from the water. The obtained oil was contained in a vial, sealed, and stored at 4°C until further analysis.

3.4 Physicochemical characteristics of the essential oils

The physicochemical properties of the essential oils determined were yield, color, odor, specific gravity, refractive index, and solubility in ethanol. The oil yield was calculated using equation 1. The color of the essential oils was observed by physical observation in daylight, and their odor was investigated by organoleptic evaluation (Evans, 2002). The specific gravity of the essential oils measured by a 10-mL pycnometer was carried out at 29°C. The pycnometer filled with oil was placed in a thermostatic bath and allowed to stabilize to the set temperature. After 30 minutes, the oil level was adjusted to a proper point on the pycnometer and stopper. Then, the pycnometer was removed from the bath, dried thoroughly, and weighed. The specific gravity was then calculated with respect to water using the formula 2 (The United States Pharmacopeial Convention, 2000). The refractive index of the essential oils was determined with an ATAGO refractometer model 3T at 29.5-29.8°C. In brief, three drops of each essential oil were applied to the refractometer. After allowing it to stand for 5 min, the reading was recorded on the display screen (The United States Pharmacopeial Convention, 2000). For the oils' solubility in ethanol, it was evaluated as directed in the method 2.8.10 of the European Pharmacopoeia (Council of Europe, 2000). The separate test was conducted based on complete solubilization of 1 mL of each essential oil in 70, 80, and 90% ethanol at 24°C. Oil volume of 1.0 mL was placed into a glass-stoppered cylinder maintained in the set temperature bath. The ethanol was added to the cylinder in increments of 0.1 mL until the solution is complete with shaking frequently and vigorously. The volume of alcohol added was recorded when a clear solution has been obtained.

The obtained physiochemical characteristics were then compared with those of the commercial oils available from TCFF and with standard specifications of the Food Chemicals Codex (FCC) (The United States Pharmacopeial Convention, 2000).

$$\text{Yield (\% v/w)} = \frac{\text{volume of oil}}{\text{weight of plant materials used for oil distillation}} \times 100 \quad (1)$$

$$\text{Specific gravity} = \frac{\text{weight of oil with 10.0 mL in volume}}{\text{weight of pure water with 10.0 mL in volume}} \quad (2)$$

3.5 TLC fingerprint of the essential oils

A TLC glass plate (20 x 20 cm) precoated with silica gel 60 GF254 (Merck, Germany) was used in this study. On the TLC plate, three drops each of standard and sample solutions were applied using a capillary tube. A TLC chamber pre-saturated with a mixture of toluene/ethyl acetate (93:7, v/v) for 20 min was employed to develop the TLC plate at room temperature. After the complete development, the TLC plate was removed and left to dry in a fume hood. The developed TLC plate was then observed under a UV lamp at 254 and 365 nm. After that, it was sprayed with a vanillin-sulfuric acid reagent and subsequently heated in a hot-air oven at 110°C for 10-15 min. Finally, a picture of the TLC chromatogram was taken in daylight.

3.6 GC-MS analysis of the essential oils

The GC-MS analysis of the essential oils was performed on a Hewlett Packard 6890 gas chromatograph coupled to an Agilent Technologies MS 5973 Model GISI3A mass spectrometer with a mass selective detector (MSD). The GC was equipped with a capillary column (Agilent: HP-5MS, HP19091 S-433; length, 30 m; internal diameter, 0.25 mm; film thickness, 0.25 µm). Helium was employed as a carrier gas at a flow rate of 0.8 mL/min. 1 µL of the sample (0.25% v/v in ethanol) was injected into the injection port set at 250°C with a split ratio of 50:1. The column temperature was initially kept at 80°C for 13 min. Then, it was ramped up to 200°C at a rate of 10°C/min and finally maintained isothermally for 2 min. Each compound was identified based on the matched mass spectrum in the spectral library.

4. Results and Discussion

4.1 Physicochemical analysis of the essential oils

Since the quality of essential oils can vary widely, analysis of the physicochemical properties which are intrinsic physical and chemical characteristics could be used to determine the quality of essential oils extracted from plants. The hydrodistilled marjoram oil (HMO) and hydrodistilled sweet basil oil (HBO) were investigated for their physicochemical properties. The comparison of the physicochemical properties of the hydrodistilled oils with those of the commercial oils was summarized in Table 1.

Table 1 The physicochemical properties of the hydrodistilled oils with a comparison to the commercial ones

Characteristics	Marjoram oil		Sweet basil oil	
	HMO[1]	TCFF[2]	HBO[3]	TCFF
1. Oil yield	0.44% v/w	nd[4]	0.20% v/w	nd[4]
2. Color	clear, light yellow	clear, colorless	clear, colorless	clear, colorless
3. Odor	pleasant	pleasant	pleasant	pleasant
4. Specific gravity	0.8922	0.9129	0.9057	0.9542
5. Refractive index	1.4708	1.4606	1.4720	1.5118
6. Solubility				
70% ethanol	1.60 mL	1.70 mL	1.70 mL	insoluble
80% ethanol	0.90 mL	0.70 mL	0.70 mL	0.90 mL
90% ethanol	0.10 mL	0.10 mL	0.30 mL	0.50 mL

[1]Hydrodistilled marjoram oil, [2]Oils obtained from Thai-China Flavours and Fragrances Industry (TCFF), [3]Hydrodistilled sweet basil oil, [4]Not determined

The oil yields of the HMO and HBO were 0.44% and 0.20% v/w, respectively. The HMO was clear and light yellow with little difference from the commercial oil (TCFF), while the HBO was clear and colorless similar to the TCFF. Both the hydrodistilled and commercial oils gave similar pleasant odors. The colors of the hydrodistilled oils were quite different from those described in the Food Chemicals Codex, which were yellow or greenish-yellow for marjoram oil and pale yellow to yellow for sweet basil oil (The United States Pharmacopeial Convention, 2010). The oil yield of the HMO was comparable to those reported from Tunisia and Greece (Baâtour et al., 2012; Komaitis, Ifanti-Papatragianni, & Melissari-Panagiotou, 1992) but had a lesser percentage than those cultivated in Egypt (2.5-3.0%) (Soliman et al., 2009). Several studies have investigated various aspects of the production process that could increase the oil yield and then come to suggestions. Seed conditioning and treatment with gibberellic acid, organic cultivation with composted manure, and bacterial inoculation were some of the production techniques that could benefit the yield (Krishnakumar & Potty, 2012).

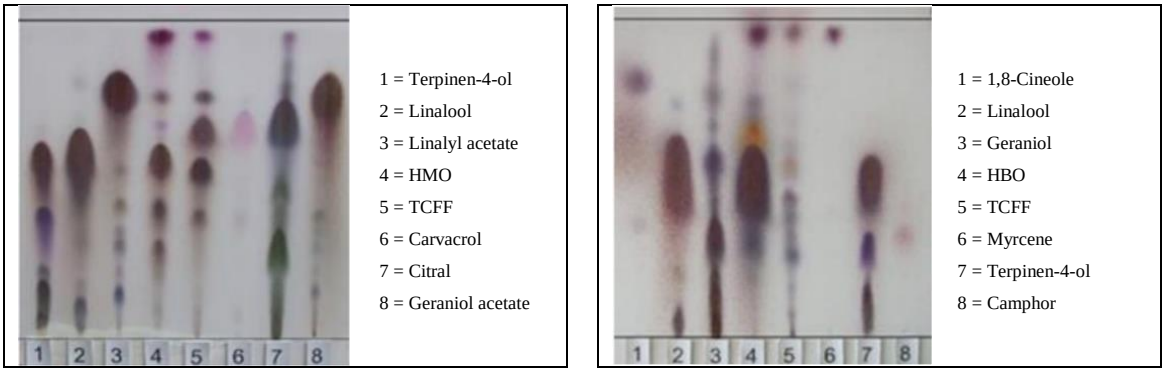
The HMO and HBO had specific gravities of 0.8922 and 0.9057, respectively, which were not much different from those of the TCFF (0.9129 and 0.9542). It was worth noting that the specific gravities of the HMO and HBO met the standard values for marjoram (0.890-0.906) and sweet basil (0.900-0.920) oils (The United States Pharmacopeial Convention, 2010). Likewise, the refractive indices of the HMO (1.4708) and HBO (1.4720) were comparable to the commercial ones (1.4606 and 1.5118). However, the refractive index

of the HBO (1.4720, measured at 29.8 ºC) was a bit lower than the standard value ranging between 1.483 and 1.493 (at 20 ºC) (The United States Pharmacopeial Convention, 2010). For the solubility in ethanol, the hydrodistilled oils exhibited the solubility similar to the TCFF oils except that the HBO was much more soluble in 70% ethanol than TCFF oil that was insoluble. This finding, indicated that the HBO contained more polar components than the TCFF oil.

4.2 TLC fingerprint

A fingerprint of essential oils is a chromatographic pattern of common chemical constituents of bioactive and/or chemical characteristics. In this study, the chemical compositions of the hydrodistilled oils in comparison to the commercial oils were preliminarily screened by TLC. The obtained TLC chromatogram (Figure 1) using a mobile phase of toluene/ethyl acetate (93:7, v/v) demonstrated that the HMO had different chemical compositions compared to the TCFF.

According to Figure 1A, the HMO exhibited 6 different spots. The presence of two minor spots with R_f values of 0.78 and 0.96 (Table 2) were observed in both the HMO and TCFF. From the physical appearance together with the R_f value, the red-brown spot with the R_f of 0.78 could be identified as linalyl acetate or geraniol acetate. Besides, the red-brown major spot of the HMO exhibited an equal R_f value of 0.56 to terpinen-4-ol and linalool implying the presence of these two monoterpenes in the HMO. For the sweet basil oils, both the HBO and TCFF gave different TLC chromatograms. As per Figure 1B, linalool was noticeably present in the HBO as compared to the standard linalool but not observed in the TCFF. This majority of linalool would thus result in a much different solubility in 70% ethanol of the HBO from that of the TCFF (Table 1). Furthermore, only myrcene (R_f 0.97) was present in both oils (Table 3), but other exhibiting compounds could not be identified from the TLC chromatogram using this mobile phase system.



A B

Figure 1 TLC chromatogram of the marjoram (A) and sweet basil (B) oils as compared with those of the TCFF oils

The results suggested that several different solvent systems should be used to make sure that the substances are identical to the R_f value and is not an absolute physical constant. Furthermore, other techniques such as GC-MS and TLC-MS should be used to unambiguously identify unknown compounds.

4.3 GC-MS analysis

The gas chromatographic analysis demonstrated that the major chemical compounds in the hydrodistilled oils were different from the commercial ones (Figure 2) (Tables 4 and 5). Of the HMO, the first three principal compounds consisted of 3-cyclohexen-1-ol (39.21%), γ-terpinene (9.00%), *trans*-sabinene hydrate (1*R*-isomer) (9.00%), while those found in the TCFF were 1,8-cineole (57.16%), linalool (17.88%), and (–)-α-terpineol (2.80%) (Table 4). Additionally, only (–)-α-terpineol and linalyl acetate were common in both oils. From the literature, there have been few reports on the presence of 3-cyclohexen-1-ol as a major compound in marjoram oil. Kang et al. (2009) reported that it contained 23.2% of 3-cyclohexen-1-ol in the marjoram oil obtained from the USA. Recently, the derivative of 3-cyclohexen-1-ol [4-methyl-1-

(1-methylethyl)-3-cyclohexen-1-ol] with an amount of 21.01% was reported as a major component in the Egyptian marjoram oil (Abdel-Meguid, Ramadan, Khater, & Radwan, 2022).

The chemical compositions of marjoram essential oils can vary widely depending on various factors such as localities, cultivars, plantation environment, and climatic conditions. For example, the oils from the shoots of marjoram grown under different cultural conditions in Tunisia gave different percentages of sabinene hydrate (Baâtour et al., 2012). These conditions resulted in the variation in the amount of *cis*- and *trans*-isomers of sabinene hydrate due to the bioconversion between them. Furthermore, marjoram grown in a hydroponic medium gave *cis*-sabinene hydrate, while the other one planted in inert sand yielded *trans*-sabinene hydrate. Gharib et al. (2008) investigated Egyptian marjoram oils and found that the relative percentages of *cis*-sabinene hydrate and terpinene-4-ol in the oils changed due to the fertilization type or level. Karousou, Efstathiou, and Lazari (2012) examined the chemical diversity of marjoram growing wild in six different localities of Cyprus. They found that two distinct oil types exist in the wild growing marjoram in Cyprus (*trans*-sabinene hydrate/terpinen-4-ol type and α-terpineol/*trans*-sabinene hydrate type). They also showed that the contents of the main compounds of marjoram essential oils obtained from cultivated plants varied depending on their origins.

For the sweet basil oils, 1,8-cineole, 1,3,7-cctatriene, *trans*-α-berganotene, β-cubebene, α-amorphene, and *epi*-bicyclosesquiphellandrene were observed in both the hydrodistilled and commercial oils. In accordance with the TLC analysis, linalool, accounting for 60.03% was dominant in the HBO followed by eugenol (19.62%). Obviously, estragole (methyl chavicol) with a large quantity of 85.79% was detected in the TCFF (Table 5). This data indicated that our hydrodistiled sweet basil oil belonged to the linalool-rich type, while that of TCFF was in the Reunion type (Akbari et al., 2018). Klimánková et al. (2008) reported linalool as a major component, ranging between 16-32%, in five cultivars of sweet basil grown in Prague. The sweet basil from Ethiopia, on the other hand, was mainly composed of estragole with 38.23% (Gebrehiwot et al., 2016). Like the marjoram essential oils, the compositions of sweet basil oils can also vary depending on those aforementioned parameters. For example, it was reported that the oil of sweet basil grown in East Turkey with a cold climate contained linalool as a major compound (Agkul, 1989), whereas estragole (methyl chavicol) was predominant in the oil obtained from Ethiopia having the tropical monsoon environment (Gebrehiwot et al., 2016).

As the result, it should be noted that the essential oil composition varies with the origin of the plant. It can be noticed in the TCFF oils imported from the Republic of China, at which the plants were grown and their oils were produced. Other factors leading to the chemical differences in these oils might be plant varieties, cultivating environment, and the extraction processes of the oils.

5. Conclusion

The physicochemical parameters and chemical compositions of the hydrodistilled marjoram and sweet basil oils were determined and compared with those of the commercial ones. The results showed that their physical properties (color, odor, specific gravity, refractive index, and solubility in ethanol) were not much different from each other. The GC-MS analysis revealed that the first three major constituents of the hydrodistilled marjoram oil were 3-cyclohexen-1-ol (39.21%), γ-terpinene (9.00%), and *trans*-sabinene hydrate (1*R*-isomer) (9.00%). On the other hand, those two commercial oil included 1,8-cineole (57.16%) and linalool (17.88%). Linalool (60.03%) and eugenol (19.62%) were mainly present in the hydrodistilled sweet basil oil, whereas estragole accounting for 85.79% was dominant in the commercial oil. Thus, our findings indicated that the essential oil of sweet basil cultivated in the highland areas of Thailand belonged to the linalool-rich group. This study exhibited that both the hydrodistilled marjoram and sweet basil oils were different from the commercial ones in terms of chemical compositions. Consequently, factors leading to the variation in the amount of the chemical components in the marjoram and sweet basil oils are worth investigating in the future. Furthermore, the study of the biological activities of the hydrodistilled oils from marjoram and sweet basil cultivated in northern Thailand should be carried out. The improvement of the cultivation of these aromatic plants to get the more qualified oils and to substitute the imported ones should be implemented in the highland area.

Generally, it can be recommended that the essential oil of sweet basil cultivated at the Royal Agricultural Station at Khun Pae can serve as a rich source of linalool.

Table 2 R_f values of components in the hydrodistilled marjoram oil (HMO) as compared to the commercial marjoram oil (TCFF) and standard compounds

Lane	Standard compounds/Oil samples	Spot	Color	UV365	R_f
1	Terpinen-4-ol	1	black	yellow-green	0.13
		2	green	yellow-green	0.21
		3	purple	purple	0.36
		4	red-brown	yellow-green	0.56
2	Linalool	1	dark-gray	purple	0.09
		2	blue	yellow-green	0.14
		3	green-brown	yellow-green	0.36
		4	red-brown	yellow-green	0.56
		5	light green	yellow-green	0.81
3	Linalyl acetate	1	blue	purple	0.13
		2	purple	yellow-green	0.21
		3	gray-blue	purple	0.28
		4	gray	yellow-green	0.34
		5	green-brown	yellow-green	0.43
		6	brown	yellow-green	0.53
		7	red-brown	yellow-green	0.79
4	Hydrodistilled marjoram oil (HMO)	1	dark brown	yellow-green	0.29
		2	dark brown	yellow-green	0.41
		3	red-brown	yellow-green	0.56
		4	red-purple	yellow-green	0.68
		5	red-brown	yellow-green	0.78
		6	violet	violet	0.96
5	Commercial marjoram oil (TCFF)	1	dark brown	-	0.39
		2	red-brown	-	0.53
		3	red-brown	-	0.66
		4	violet	-	0.71
		5	red-brown	-	0.78
		6	violet	violet	0.96
6	Carvacrol	1	red-brown (not clear)	yellow-green	0.38
		2	pink-violet	-	0.67
7	Citral	1	green	-	0.26
		2	green	-	0.36
		3	green	-	0.46
		4	blue	-	0.68
		5	gray blue	-	0.79
		6	green	-	0.87
		7	violet	violet	0.97
8	Geraniol acetate	1	light blue	-	0.14
		2	brown	-	0.21
		3	light blue	-	0.29
		4	blue	-	0.34
		5	yellowish green	-	0.39
		6	red-brown	-	0.76

Table 3 R_f values of components in the hydrodistilled sweet basil oil (HBO) as compared to the commercial marjoram oil (TCFF) and standard compounds

Lane	Standard compounds/Oil samples	Spot	Color	UV365	R_f
1	1,8-Cineole	1	violet	-	0.33
		2	purple	-	0.79
2	Linalool	1	red-brown	-	0.03
		2	brown	-	0.18
		3	red-brown	-	0.59
		4	blue-green	-	0.71
3	Geraniol	1	red	-	0.04
		2	red-brown	-	0.12
		3	green	-	0.16
		4	red-brown	-	0.29
		5	black	-	0.39
		6	purple	-	0.52
		7	blue	-	0.66
		8	gray-blue	yellow	0.75
		9	purple	yellow	0.88
4	Hydrodistilled sweet basil oil (HBO)	1	blue	-	0.27
		2	red-brown	-	0.57
		3	yellow-orange	-	0.62
		4	blue-green	yellow	0.69
		5	purple	yellow	0.74
		6	violet	violet	0.97
5	Commercial sweet basil oil (TCFF)	1	blue	-	0.04
		2	blue	pink-orange	0.11
		3	blue	-	0.24
		4	violet	light purple	0.30
		5	blue	-	0.37
		6	purple	-	0.43
		7	yellow-orange	yellow	0.53
		8	purple (not clear)	-	0.59
		9	purple (not clear)	-	0.67
		10	purple	-	0.79
		11	violet	violet	0.97
6	Myrcene	1	violet	violet	0.97
7	Terpinen-4-ol	1	red-brown	-	0.06
		2	purple	purple	0.26
		3	red-brown	-	0.45
8	Camphor	1	red-brown	yellow-green	0.29

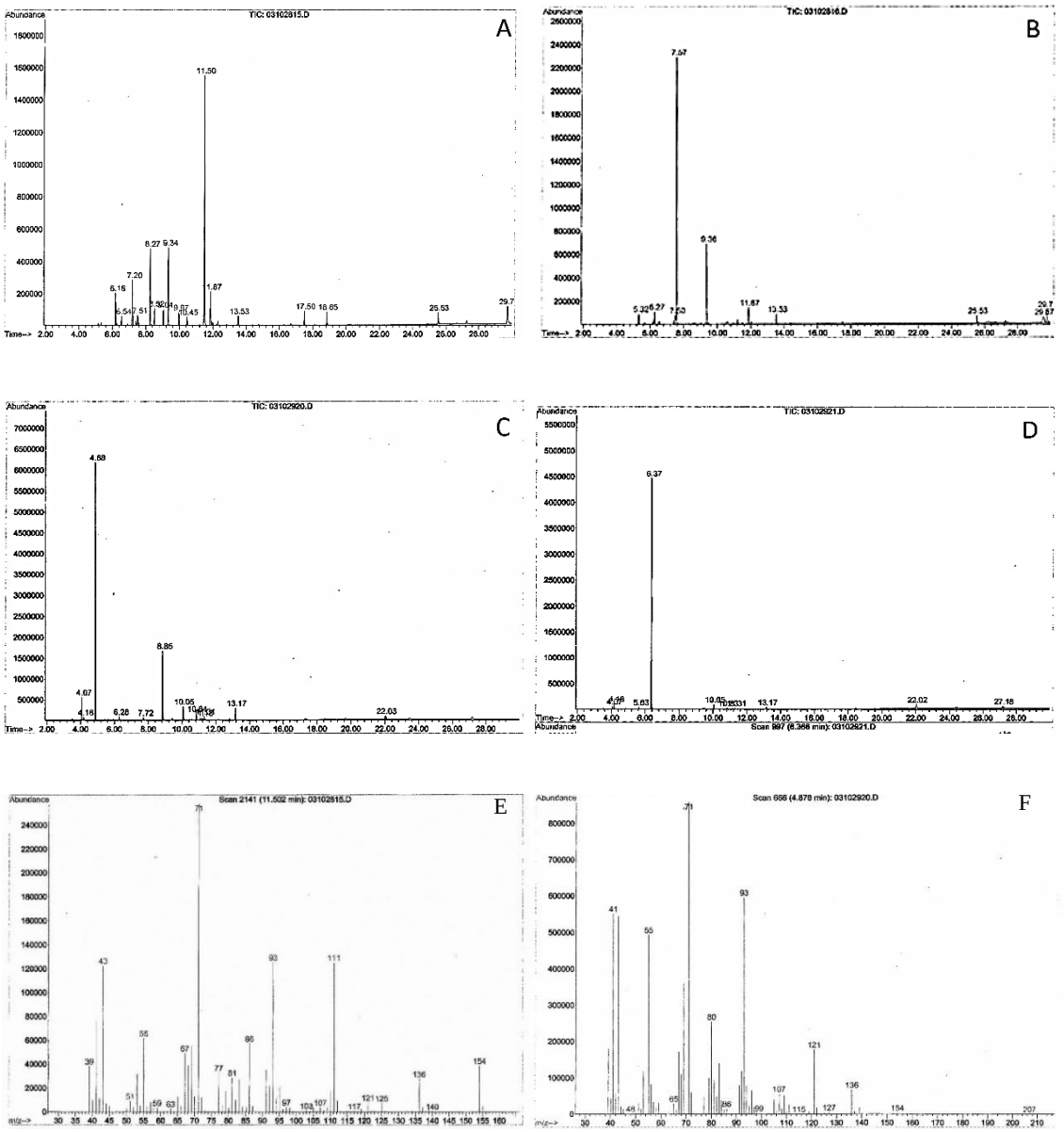


Figure 2 The gas chromatograms of marjoram oils (A, the hydrodistilled essential oil and B, TCFF oil) and sweet basil oils (C, the hydrodistilled essential oil and D, TCFF oil) together with the representative mass spectra of the hydrodistilled majoram (E) and sweet basil (F) oils demonstrating the peaks of 3-cyclohexen-1-ol and linalool at 11.50 and 4.88 min, respectively

Table 4 Chemical constituents of the hydrodistilled marjoram oil (HMO) as compared to the commercial marjoram oil (TCFF)

No.	Retention time (min)	Compounds	Relative quantity (%)	
			HMO	TCFF
1	5.32	α-Pinene	-	1.871
2	6.18	Sabinene	4.004	-
3	6.27	β-Pinene	-	2.541
4	6.55	Myrcene	0.900	-
5	7.20	α-Terpine	6.034	-
6	7.50	Limonene	-	1.824
7	7.52	Sabinene (1*R*-isomer)	0.900	-
8	7.57	1,8-Cineole	-	57.16
9	8.27	γ-Terpinene	9.000	-
10	8.52	*trans*-Sabinene hydrate	1.000	-
11	9.04	Terpinolene	1.000	-
12	9.34	*trans*-Sabinene hydrate (1*R*-isomer)	9.000	-
13	9.35	Linalool	-	17.877
14	9.97	2-Cyclohexen-1-ol	0.800	-
15	10.45	1-Terpineol	0.500	-
16	11.50	3-Cyclohexen-1-ol	39.210	-
17	11.86	(–)-α-Terpineol	4.000	2.800
18	13.53	Linalyl acetate	0.500	1.824
19	17.50	β-Caryophyllene	0.900	-
20	18.85	Bicyclogermacrene	0.800	-
21	25.53	Benz[a]acridine	-	1.510
22	29.57	8-Nitro-2,3-dimethylpyrrolo	1.000	-

Table 5 Chemical constituents of the hydrodistilled sweet basil oil (HBO) as compared to the commercial sweet basil oil (TCFF)

No.	Retention time (min)	Compounds	Relative quantity (%)	
			HBO	TCFF
1	4.07	1,8-Cineole	5.101	1.275
2	4.18	1,3,7-Octatriene	0.709	1.826
3	4.88	Linalool	60.029	-
4	5.63	L-Camphor	-	0.917
5	6.28	Linalyl propionate	0.938	-
6	6.37	Estragole	-	85.789
7	7.72	(–)-Bornyl acetate	0.697	-
8	8.85	Eugenol	19.620	-
9	10.05	*trans*-α-Berganotene	3.719	2.088
10	10.83	β-Cubebene	1.927	0.648
11	11.18	*trans*-γ-Bisabolene	0.901	-
12	11.31	α-Amorphene	1.095	0.577
13	13.17	epi-Bicyclosesquiphellandrene	3.540	1.134
14	22.023	8-Nitro-2,3-dimethylpyrrole	-	2.873
15	22.025	4*H*-1-Benzopyran-4-one	1.724	-
16	27.18	Benz[a]acridine	-	2.872

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