Evaluation of the effect of extraction method on chemical composition and antioxidant activity of

The Lime Essential Oil

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BACKGROUND

Lime *Citrus aurantifolia* (Christm. et Panz.) Swing has a smooth, thin skin, is greenish-yellow in color, and has a tiny neck, a flat base, and a small nipple at the apex gaining increasing recognition for its economic and medicinal value (Lin et al, 2019). Lime peels, often regarded as waste, constitute approximately 27% of the raw processed fruit and represent a potential source of valuable by-products. (Russo et al, 2021). The essential oils extracted from lime peels are widely utilized in the perfume, food, and beverage industries, as well as for medicinal purposes, due to their antioxidant properties, which are primarily attributed to compounds such as terpenes.

OBJECTIVES

This study investigated the effect of extraction methods (water and steam distillation, supercritical CO_2 extraction, and microwave extraction) on the chemical composition and antioxidant activity of lime peel essential oil.

Lime

Citrus aurantifolia (Christm. et Panz.) Swing

Lime Peel

27%

of the raw processed fruit often regarded as waste.

Value Added

METHODS

Chemical Composition

GC-MS

analyzed using gas chromatography-mass spectrometry (GC-MS) with a Nexis GC-2030 model equipped with a DB-5MS column (Babarinde et al., 2021).

Antioxidant Activites

- **DPPH** radical scavenging activity described by **Zhu et al. (2006)**
- **ABTS** free radical scavenging activity modified from **Re et al. (1999).**

RESULTS & DISCUSSIONS

Yield and Characteristics

Figure 1 Yield and characteristics of lime essential oil obtained from water and steam distillation, supercritical CO_2 extraction and microwave extraction.

Lime essential oils were extracted using water and steam distillation (WS), supercritical CO_2 extraction (SFE), and microwave extraction (ME). The yields of essential oils obtained through these methods are presented in Figure 1. **The highest yield was achieved using SFE (3.03±0.48%)**, followed by WS (1.21±0.05%) and ME (0.45±0.12%). The extracted lime essential oils exhibited a clear to dark brown color and a characteristic aromatic odor.

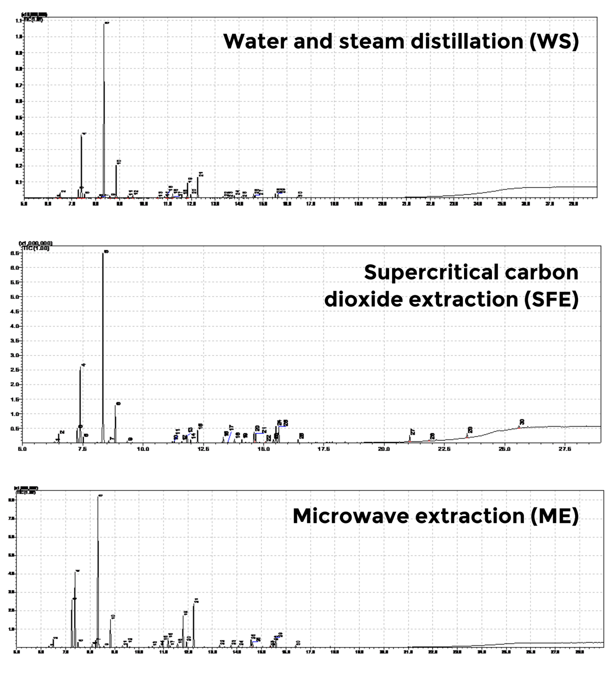


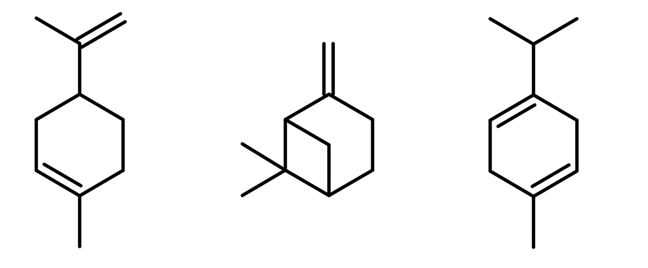
Figure 2 The GC Chromatograms of lime essentials oils.

The composition of lime essentials oils was analyzed by GC-MS. 40 compounds of the essential oils were identified. The essential oils samples were rich in monoterpenes (Table 1). **The major components of the lime essential oils from all extraction methods were similar, with D-Limonene, γ-terpinene, and β-pinene** being the main identified compounds, these results agreed with the previous work of Lin et al. (2019). D-limonene is the dominant compound with concentrations of 43.55±0.76%, 42.74±0.37%, and 33.06±1.49 in WS, SFE, and ME, respectively.

Table 1 The chemical composition of lime essential oils identified using GC-MS.

NO	Possible compound	Retention Time	KI Index	% area WS	% area SFE	% area ME
1	α-Thujene	6.327	930	0.31±0.03	0.31±0.00	-0.26±0.07
2	α-Pinene	6.501	939	1.75±0.11	1.93±0.01	1.71±0.09
3	Camphene	6.840	954	-	-	
4	Sabinene	7.266	975	2.41±0.01	3.06±0.02	6.84±3.55
5	β-Pinene	7.394	979	17.66±0.71	16.51±0.12	15.84±0.04
6	β-Myrcene	7.520	994	1.11±0.02	1.21±0.00	1.00±0.12
7	α-Terpinene	8.101	1017	0.48±0.03	-	0.50±0.17
8	p-Cymene	8.244	1024	0.28±0.01	-	1.16±0.41
9	D-Limonene	8.336	1025	43.55±0.76	42.74±0.37	33.06±1.49
10	(E)-β-Ocimene	8.583	1050	0.42±0.03	0.54±0.01	0.31±0.02
11	γ-Terpinene	8.849	1065	8.52±0.48	7.50±0.11	5.77±0.35
12	α-Terpinolene	9.344	1091	0.59±0.05	0.39±0.01	0.40±0.02
13	Linalool	9.548	1096	0.64±0.07	-	0.89±0.10
14	Fenchyl alcohol	9.956	1116	-	-	-
15	α-Phellandren-8-ol	10.794	1170	-	-	-
16	Isoneral	10.572	1160	0.21±0.04	-	0.25±0.02
17	(Z)-isocitral	10.879	1164	0.36±0.05	-	0.41±0.03
18	Terpinen-4-ol	11.000	1177	1.46±0.06	-	1.35±0.22
19	α-Terpineol	11.216	1192	1.73±0.03	0.36±0.01	2.10±0.25
20	*n*-Decanal	11.296	1201	0.34±0.02	0.28±0.01	0.49±0.11
21	Citronellol	11.565	1225	-	-	
22	Nerol	11.605	1230	1.03±0.23	0.35±0.02	1.00±0.23
23	Neral	11.833	1244	4.51±0.53	1.49±0.04	6.74±0.55
24	(E)-Geraniol	11.972	1257	1.12±0.23	0.68±0.04	1.74±0.81
25	(E)-Geranial	12.264	1273	5.92±0.64	2.58±0.05	9.39±0.77
26	δ-Elemene	13.325	1338	0.38±0.05	1.11±0.02	0.51±0.05
27	Neryl Acetate	13.527	1361	0.25±0.02	0.26±0.00	
28	Geranyl acetate	13.798	1381	0.83±0.08	0.75±0.02	0.75±0.44
29	β-Elemene	14.100	1390	0.29±0.04	0.73±0.03	0.41±0.04
30	β-Caryophyllene	14.604	1426	0.91±0.09	2.63±0.03	1.70±0.21
31	(E)-α-Bergamotene	14.672	1434	0.59±0.10	2.07±0.04	1.13±0.10
32	α-Humulene	15.083	1454	-	0.27±0.01	
33	Germacrene D	15.401	1485	-	0.88±0.02	0.30±0.04
34	α-Farnesene	15.514	1505	1.08±0.11	3.39±0.02	1.48±0.10
35	β-Bisabolene	15.629	1505	1.01±0.16	3.56±0.02	2.08±0.12
36	Germacrene B	16.433	1561	0.25±0.01	0.77±0.01	0.45±0.03
37	Citraptene	21.055	1916	-	1.51±0.21	-
38	Bergaptene	21.864	2048	-	0.34±0.04	-
39	Isopimpinellin	23.435	2227	-	1.02±0.13	-
40	Oxypeucedanin	25.585	2765	-	0.74±0.05	-

Chemical Composition



D-Limonene β-Pinene γ-Terpinene

Lime Essential Oils Major components

Antioxidant activities

Table 2 Antioxidant activity of lime essential oils from different extraction methods and its main chemical composition using DPPH and ABTS assays.

Sample	DPPH assay IC_{50} (mg/ml)	ABTS assay IC_{50} (mg/ml)
Lime oil (WS)	151.66±1.59	516.93±2.13
Lime oil (ME)	76.73±5.26	455.41±17.38
Lime oil (SFE)	8.45±0.19	133.82±0.61
γ- Terpinene	42.20±3.04	205.27±1.48
β- Pinene	119.18±4.61	>1,000mg/ml (33.30%±0.36 at conc. 1,000 mg/ml)
Limonene	374.61±3.72	>1,000mg/ml (10.30%±0.31 at conc. 1,000 mg/ml)

The antioxidant capacities of lime essential oil and its main components were evaluated using the DPPH and ABTS radical-scavenging assays. Table 2 presents the IC_{50} values, which quantify the concentrations required to neutralize 50% of free radicals, as a measure of antioxidant activity. **The essential oil obtained via supercritical CO_2 extraction (SFE) demonstrated the highest activity in both assays**, with IC_{50} values of 8.45±0.19 mg/mL for DPPH and 133.82±0.61 mg/mL for ABTS, followed by the oils extracted using microwave extraction (ME) and water and steam distillation (WS). Among the constituents, γ-terpinene was identified as a significant contributor to the antioxidant activity, with IC_{50} values of 42.20±3.04 mg/mL in the DPPH assay and 205.27±1.48 mg/mL in the ABTS assay.

Conclusions

In conclusion, lime essential oil was successfully extracted from the peel of *Citrus aurantifolia* (Christm. et Panz.) Swing using supercritical carbon dioxide extraction (SFE), achieving the highest yield of 3.03±0.48%. The oil is rich in limonene, which is its major component, followed by β-pinene and γ-terpinene. Additionally, antioxidant activity evaluation using DPPH and ABTS assay demonstrated that lime essential oil obtained through SFE exhibited the highest antioxidant activity, with γ-terpinene contributing significantly to this activity.

Acknowledgements

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