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Research Article

The influence of different types of mulch on the yield and content of bioactive compounds of immortelle

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Abstract

Immortelle (*Helichrysum italicum*) is a perennial Mediterranean plant known for its medicinal and aromatic properties. Various biotic and abiotic factors influence the yield and quality of immortelle. Mulching is a sustainable agricultural practice that conserves soil moisture, prevents weed growth, and regulates soil temperature. The aim of this research was to examine the influence of different types of mulch on the yield and quality of immortelles. The study had three types of mulch (alfalfa, straw, and perennial mulch) and a control (no mulch). The research mulches significantly affected the yield, essential oil (EO) content and content of bioactive compounds. The highest yield of inflorescences was recorded with alfalfa mulch (67.5 g^{-1}), while the control had the lowest yield (37.3 g^{-1}). The EO content was highest in treatments with straw and control (0.33 mL 100 g^{-1}), whereas the lowest content was observed with alfalfa mulch (0.28 mL/100 g). The total phenolic content reached its maximum in the control variant (75.1 mg GAE g^{1}), while perennial mulch resulted in the highest flavonoid content (20.4 mg CAE g^{-1}). A total of 63 constituents were identified in the EO. The largest share was: neryl acetate, α-curcumen, italidione, α-pinene and D-limonene, and others in smaller quantities.

Keywords: antioxidant activity; essential oil; immortelle; mulch; neryl acetate; quality; yield

Introduction

Immortelle (*Helichrysum italicum*) is a perennial species with medicinal, spicy, and aromatic properties. It is used in traditional medicine as a diuretic, choleretic, and expectorant (Dănăilă-Guidea *et al.*, 2022; Kunc *et al.*, 2022). Nowadays, it is mainly used in high-end skin care cosmetics due to its regenerative properties and in the production of luxury perfumes (Hellivan, 2009). Also, much research has been directed towards studying antioxidant, anti-inflammatory, antiviral, antimicrobial and antiproliferative effects and their positive impact on humans (Ninčević *et al.*, 2019). Immortelle's medicinal properties arise from its essential oils (EOs) specific chemical composition. The primary constituents in immortelle EO include neryl acetate, γ-curcumene, (+)-limonene, neryl propionate, α-pinene, ar-curcumene, italidione, nerol, italicene, linalool and other constituents in small amounts (Tisserand and Young, 2013; Erbaş *et al.*, 2023; Furlan and Bren, 2023).

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The cultivation of immortelle is traditionally associated with the Mediterranean region. However, it has recently gained popularity in the continental climate of Bosnia and Herzegovina (B&H). Although these plant species have been reported in the B&H market, no official data regarding their cultivated areas, cultivation practices, or quality is available. The yield and quality of immortelle are affected by various biotic and abiotic factors. Previous research has shown that abiotic factors affect the yield and synthesis of bioactive compounds in medicinal plants (Gavrić *et al.*, 2024). Also, the type of agrotechnical practices used can significantly affect the yield and quality of these plants (Šakonjić *et al.*, 2023).

In recent years, mulching has become helpful in sustainable agriculture due to its various benefits. Mulching prevents water evaporation, which is especially important in dry regions (Zribi *et al.*, 2015; El-Beltagi *et al.*, 2022) . In addition, it prevents the growth of weeds (Hasanvand *et al.*, 2023). It creates a barrier on the soil surface that prevents the development of weeds. Mulching regulates soil temperatures (Palada *et al.*, 2000; Zengin and Sabir, 2022; Hasanvand *et al.*, 2023;). For example, black plastic mulch warms the soil, while the straw underneath insulates it from heat. Some types of mulch (straw, leaves, hay, and sawdust) decompose, adding nutrients to the soil and improving its fertility (Iqbal *et al.*, 2020). Furthermore, mulch protects the soil from erosion (Šakonjić *et al.*, 2023). Therefore, it is evident that mulching changes environmental factors that can significantly affect the yield and quality of immortelle, which has not yet been clarified in detail. Given that immortelle has not been sufficiently researched in the continental agroecological conditions of B&H and that mulch positively affects cultivated crops, research in this direction becomes necessary. The aim of this study was to investigate the impact of various types of mulch materials on the yield and quality of immortelle EO.

Materials and Methods

Field experiments and experimental design

The research was conducted in the Faculty of Agriculture and Food Sciences Sarajevo experimental field in Butmir in 2024. The field experiment was designed using a split-plot plan with a basic plot area of 18 m^2. It included four different mulch treatments: perennial (agritela), straw mulch, alfalfa hay mulch, and control (no mulch). The immortelle plantation was one year old (first year of cultivation). An analysis of the soil's chemical fertility in the research area was conducted prior to the experiment setup. Before initiating the field experiment, soil samples were collected from the upper horizon (up to 20 cm deep). The collected samples were analysed to determine the soil pH (ISO 10390, 1994), organic matter content (ISO 14235, 1998), and the levels of P_2O_5 and K_2O (Egnér *et al.*, 1960). The analysis revealed the following chemical properties of the soil: a pH of 6.71, 3.3% organic matter, P_2O_5 content of 15.0 mg 100 g^{-1}, and K_2O content of 20.0 mg 100 g^{-1}. The soil was classified as an alluvial type. The plant height was measured just prior to harvesting the immortelle. Following the field experiment, research continued in the laboratories, where several parameters were assessed.

Materials for analysis

Inflorescence samples (flowers with 5-6 cm stems) were collected to estimate the yield and content of bioactive compounds from immortelle. Ten samplings were conducted in each treatment (agritela, straw mulch, alfalfa mulch, and control). The samples were collected in the blooming stage on June 20, 2024. After collection, the samples were dried in low-light conditions for 10 days at room temperature. Once dried, the samples were weighed, and the yield per plant was recorded. The total phenolic content, total flavonoid content, antioxidant activity, and EO content were then analysed in the inflorescence samples.

Weather conditions

For the analysis of weather conditions, data collected from the meteorological station of the Federal Hydrometeorological Institute of B&H were used. The measuring station was located 10 km from the experiment. This study focused on average monthly air temperatures and precipitation amounts in 2024. As indicated in Figure 1, average air temperatures during the research period varied considerably. The lowest average monthly temperature was recorded in January (1.7 °C), while the highest was observed in August (24.3 °C). Precipitation levels during the study were generally high, except in the summer months. Specifically, the least rainfall occurred in July, totalling 33 mm, and in August, with only 18 mm.

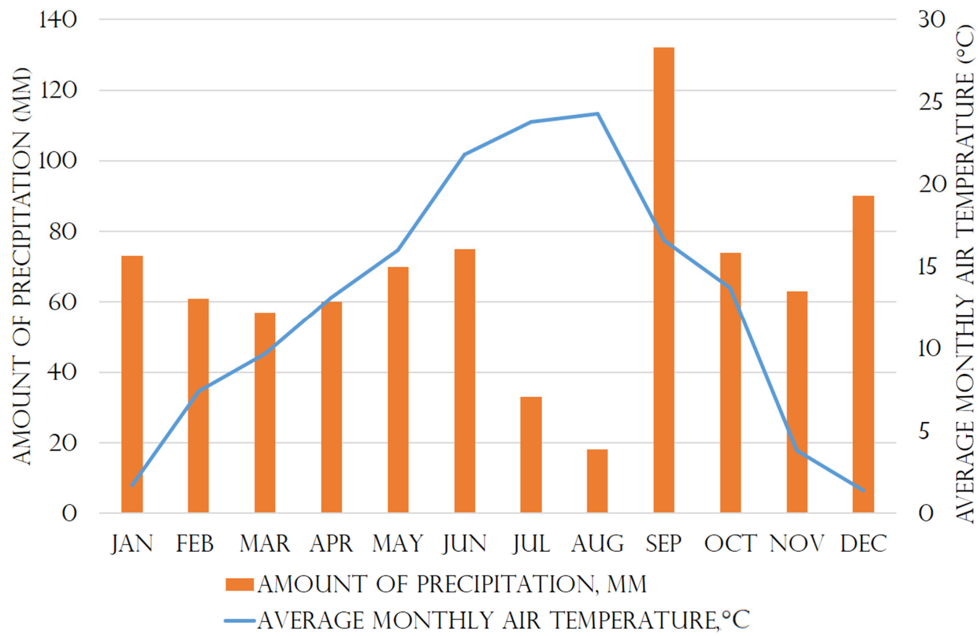


Figure 1. Average monthly temperature and monthly rainfall in 2024

Extraction of bioactive compounds

Before extracting the bioactive compounds, the immortelle inflorescences were ground into fine particles. The extraction process was conducted as follows: 1 g of the ground sample was weighed and transferred to a glass container. The sample was then immersed in 40 mL of 60% ethanol. After a 'cold' extraction lasting 24 hours, the samples were filtered through quantitative filter paper (blue strip).

Determination of total phenols

The total phenol content of the ethanol extracts was determined by using the Folin-Ciocalteu reagent, according to the procedure described by Gavrić *et al.* (2023). A sample of 0.4 mL was added to the Eppendorf tubes using a pipette. After that, 0.25 mL of Folin-Ciocalteu reagent was added (diluted 1:2 immediately before use). After 3 minutes, 0.75 mL of saturated Na_2CO_3 solution was added. Finally, 3 mL of distilled water was added. The contents prepared in this way were left at room temperature for 120 minutes until a stable blue colour developed. After developing the colour at room temperature, the absorbance is read at 765 nm. Total phenol content is expressed in mg of gallic acid equivalents (GAE).

Determination of flavonoids

The content of total flavonoids in the extract was determined by a modified spectrophotometric method (Šakonjić *et al.*, 2023). Briefly: 0.5 mL of extract, blank or standard and 2 mL of distilled water were added to a 5 mL volumetric flask. Then 0.15 mL of 5% $NaNO_2$ was added to the flask, and after 6 minutes, 0.15 mL of 10% $AlCl_3$ was added. After 6 minutes, 1 mL of 1M NaOH was added, and the flask was filled to the mark with distilled water. After 120 minutes of incubation, the absorbance was measured at a wavelength of 510 nm. The total flavonoid content was expressed in mg of catechin equivalents (CAE).

Determination of antioxidant activity

Antioxidant activity was determined by the spectrophotometric method (Benzie & Strain, 1996). First, FRAP (ferric reducing antioxidant power) reagent was prepared by mixing 25 mL of 300 mM acetate buffer, 2.5 mL of 10 mM TPTZ solution and 2.5 mL of 20 mM $FeCl_3 \times 6H_2O$ (Iron (III) chloride hexahydrate), which was heated to 37 °C. A TPTZ solution, with a concentration of 10 mM, was obtained by dissolving 159.4 mg of TPTZ in 50 mL of hydrochloric acid (40 mM HCl). After incubation, the absorbance was measured at a wavelength of 595 nm. The absorbance of a standard $FeSO_4$ solution was used as a reference measurement to determine the antioxidant activity of the sample.

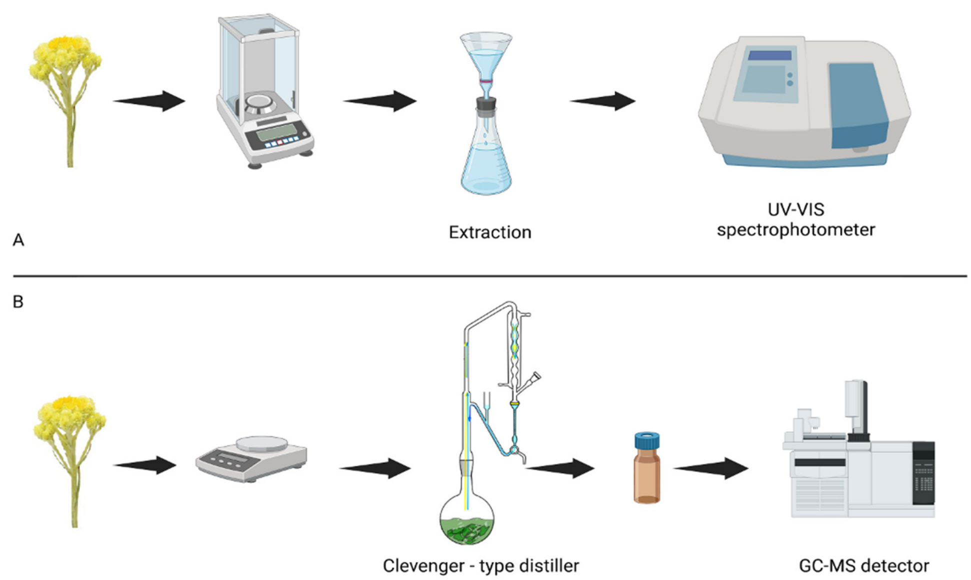


Figure 2. Analysis of total phenolics, flavonoids, and antioxidant activity (A); Isolation and determination of EO constituents (B). Figure created on BioRender.com

Isolation and determination of Essential Oil (EO)

The EO was isolated from immortelle samples using a Clevenger-type apparatus (Clevenger, 1928). First, 50 grams of dried samples were weighed and placed into a 1,000 mL volumetric flask. Then, 500 mL of distilled water was added to the flask, and the distillation process commenced. This hydrodistillation lasted for 2 hours. After the distillation process, the volume of the isolated EO was measured. The extracted EO were then dried using anhydrous sodium sulfate, transferred into 2 mL dark vials, and stored in a refrigerator until analysis.

Gas chromatography/mass spectrometry analysis

The EO constituents were analysed using a gas chromatograph with an autosampler (Agilent Technologies 7820 A) featuring a capillary column (Agilent HP5-ms ultra-inert, 30 m × 320 μm × 0.25 μm) connected to an Agilent Technologies Mass Selective Detector (MSD-5977E). The temperature program was set as follows: it started at 60 °C with a 1-minute hold. The temperature then increased at a rate of 3 °C per minute until it reached 246 °C, with no holding time. After that, the temperature was further raised at a rate of 10 °C per minute until it reached 280 °C. The sample injection was performed using a volume of 1 μL in splitless injection mode, at a temperature of 220 °C and a pressure of 58.328 kPa. Helium with a purity of 99.9995% was used as the carrier gas

Statistical methods

All experimental measurements were statistically analysed using ANOVA at a significance level of 0.05. The statistical analyses were conducted using SPSS 22.0 software (IBM, Armonk, New York, USA).

Results and Discussion

The results of the mulch type's influence on immortelle's researched traits are presented in Tables 1 and 2.

Table 1. Mulch type effect on research traits of immortelle

Type of mulch	Plant height (cm)	Yield (g per plant)	Essential oil (mL per 100 g)
Perennial	35.69 ± 3.43a	45.0 ± 16.8ab	0.29
Stra w	31.17 ± 3.5b	53.9 ± 25.1ab	0.33
Alfaafla	35.67 ± 7.8a	67.5 ± 25.5a	0.28
No mulch	34.4 ± 3.24a	37.3 ± 16.4b	0.33
Average	33.23	50.9	0.31

Different letters indicate significant differences between mulch treatments at the 0.05 level

Table 2. Mulch type effect on the content of bioactive components

Type of mulch	Total phenols contents mg GAE g^{-1}	Total flavonoid contents mg CAE g^{-}	Antioxidant activity mM Fe2+ / g
Perennial	64.5 ± 11.8b	20.4 ± 0.6a	35.9 ± 2.19ns
Straw	64.5 ± 2.75b	17.4 ± 0.55b	32.9 ± 1.37ns
Alfafla	66.5 ± 2.45b	18.1 ± 0.44b	33.7 ± 2.30ns
No mulch	75.1 ± 4.44a	19.2 ± 0.82ab	37.5 ± 1.12ns
Average	67.7	19.7	35.0

Different letters indicate significant differences between mulch treatments at the 0.05 level

The straw and alfalfa mulch treatments resulted in the highest average plant height (35.6 cm and 35.6 cm), while the lowest height was recorded in the perennial mulch treatment (31.1 cm). The inflorescence yield per plant was the highest in the alfalfa treatment (67.5 g per plant), while the lowest was recorded in the control treatment (37.3 g per plant). The results of the study suggest that yield was low in all mulch treatments. The yield is below the values commonly found in the literature and lower than those reported by other authors (Jovović *et al.*, 2016). The cause of the low yield is the young planting. That is, the research was conducted in the first year of farming. However, many researchers believe immortelle has a high yield after the third year of cultivation (Gadžo *et al.*, 2017). Despite the low yields, this study showed that mulch can affect yield. Variations in yield are likely due to differences in soil warming and fertility, which affect the growth of immortelle. This aligns with prior research indicating that soil with black mulch, such as perennial mulch, was warmer than the control group, while soil covered with straw was cooler than the control group without mulch (Palada et al., 2000; Ning et al., *2021;)*. On the other hand, alfalfa mulch contains a high amount of nutrients and positively affects plant growth (Wiens *et al.*, 2006).

The analysis of EO content reveals minor variations (Table 1). The EO content in immortelle inflorescences varied from 0.29 mL/100 g (perennial mulch) to 0.33 mL/100 g (straw and control). Although the research included various treatments that altered microenvironmental conditions, there was no significant change in the EO content of immortelle inflorescences. The lack of mulch effect could have been due to other factors. Namely, it is known that the EO content in a plant depends on various factors, including the place of cultivation, altitude, climate, cultivar, harvest time, and the locations and hours of harvest, etc. (Moghaddam *et al.*, 2023; Bahadırlı *et al.*, 2024). However, our research aligns with previous research that showed that EO varied from 0.02 to 0.78% (Aćimović *et al.*, 2022).

Different mulch types significantly influenced the bioactive components' content (Table 2). The highest content was recorded in the control variant (75.1 mg/g GAE), followed by the variants with alfalfa (75.1 mg/g GAE), perennial mulch and straw (64.5 and 64.5 mg/g GAE). Flavonoid content significantly depended on the type of mulch used, with values ranging from 17.4 mg/g CAE (straw) to 20.4 mg/g CAE (perennial mulch). Antioxidant activity varied from 33.7 (alfalfa) to 37.5 mM Fe2+/g (control). These results correlate with earlier research by Šakonjić *et al.* (2023), where it was reported that mulching affects the content of bioactive compounds in medicinal plants. These authors state that mulching changes environmental conditions, affecting secondary metabolite synthesis. Namely, it has long been known that, in addition to plant genes and agricultural practices, the environment significantly influences the synthesis of phenolic compounds and antioxidant activity (Gavrić *et al.*, 2023). Regardless, this research found that immortelle has high total phenols and flavonoid content and antioxidant activity. These findings align with previous studies highlighting immortelle as a supercritical extract, which can serve as a valuable natural antioxidant in both the food and cosmetic industries (Hellivan, 2009; Ninčević et al., 2019).

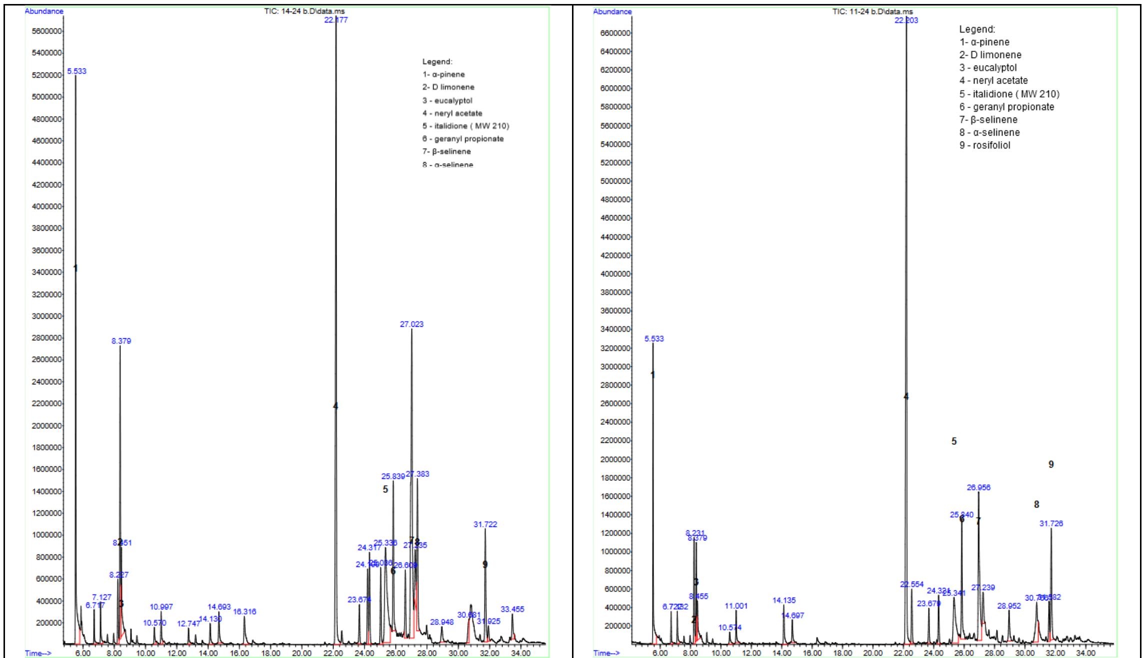
The results presented in Table 3 indicate a positive correlation among several research traits. A very strong positive correlation existed between total phenolic content and antioxidant activity (r = 0.994). A positive correlation was also observed between total phenolic content and antioxidant activity (r = 0.752) and between antioxidant capacity and flavonoid content (r = 0.735). Conversely, yield negatively correlated with total phenolic content (r = -0.615) and antioxidant capacity (r = -0.549). The results of this study are consistent with earlier studies that observed a positive correlation between antioxidant activity, total phenol content and total flavonoids (Gavrić *et al.*, 2023). Majkowska-Gadomska *et al.* (2017) believe that the high antioxidant value of medicinal herbs is due to provitamin A, vitamin C, organic acids, anthocyanins, and phenolic compounds. However, immortelle has been found to have a high content of bioactive compounds, which can be used as a source of antioxidants to neutralise free radicals that can negatively affect human health.

Table 3. The correlation between yield, total phenols content, total flavonoids content and antioxidant activity

Correlated traits	Yield	Total phenols content	Total flavonoids content	Antioxidant activity
Yield	1			
Total phenols content	-0.615*	1		
Total flavonoids	-0.327	0.395	1	
Antioxidant activity	-0.549	0.752**	0.735**	1

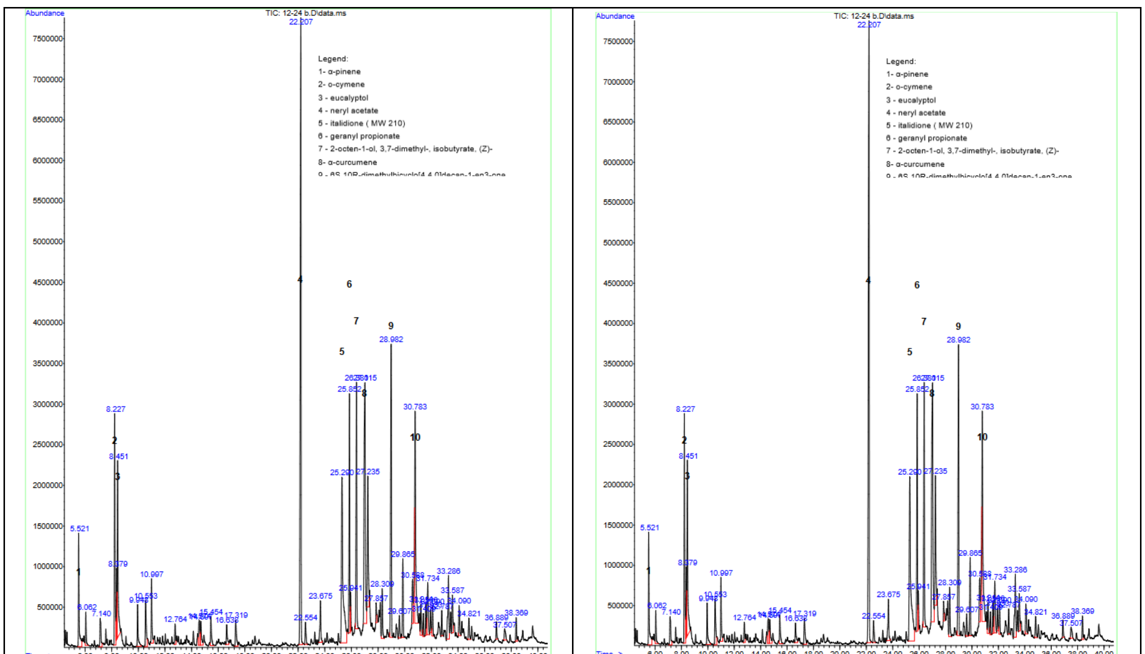
**. Correlation is significant at the 0.01 level; *. Correlation is significant at the 0.05 level

The quality of the EO of aromatic plants, including immortelle, mostly depends on the content of the constituents. Our results indicate that a total of 63 different constituents were isolated from immortelle EO with their composition influenced by the application of various mulch types (Figure 3A, 3B, 4A, 4B, and Table 4). The constituents identified in the EO included neryl acetate (18.27-32.29%), α-curcumene (7.42-14.81%), italidione (4.18-10.96%), α-pinene (2.62-11.43%), and D-limonene (1.41-6.06%), along with several other compounds present in smaller amounts. These results align with previous research (Tisserand and Young, 2013; Erbaş *et al.*, 2023; Furlan and Bren, 2023; Bahadırlı *et al.*, 2024). Our findings demonstrate that neryl acetate constitutes the highest proportion of the EO. Previous studies have shown that samples rich in neryl acetate and γ-curcumene exhibit more pronounced antimicrobial activity and stronger antioxidant potential compared to those rich in α-pinene and γ-curcumene (Mollova *et al.*, 2020).



(A) (B)

Figure 3. Chromatogram of immortelle EO: alfalfa (A) and strow (B)



(A) (B)

Figure 4. Chromatogram of immortelle EO:perennial (A) and control (B)

Table 4. Mulch type effect on essential oil chemical composition (%)

No	Constituents name	RI	Mulch			
			Alfalfa	Straw	Perennial	Control
1	α-pinene	929	11.438	9.029	8.284	2.624
2	d-camphene	951	0.326	nd	nd	nd
3	tuja-2,4(109-diene	956	nd	nd	nd	0.666
4	β-pinene	979	0.688	0.461	0.904	0.963
5	β-myrcene	991	0.927	1.050	1.137	nd
6	α-phellandrene	1005	0.154	0.158	0.183	nd
7	α-terpinene	1017	0.242	0.243	0.307	nd
8	o-cymene	1022	1.349	1.714	3.223	4.852
9	D limonene	1030	6.068	2.890	3.243	1.401
10	eucalyptol	1032	2.531	3.044	1.706	4.295
11	cis-ocimene	1038	0.348	0.377	0.427	nd
12	γ-terpinene	1060	0.191	0.168	nd	nd
13	cis-linalool oxide	1074	0.482	nd	nd	1.073
14	trans-linalool oxide	1086	nd	nd	nd	1.437
15	α-terpinolene	1088	nd	0.495	0.653	nd
16	linalool	1099	0.844	1.971	1.173	1.403
17	α-campholenal	1125	nd	nd	nd	0.240
18	L-camphor	1142	0.475	nd	nd	nd
19	trans-verbenol	1144	nd	nd	nd	0.515
20	2-methylbutyl angelate	1146	0.274	0.167	nd	nd
21	borneol	1166	nd	nd	nd	0.212
22	endo-borneol	1167	0.102	nd	nd	nd
23	terpinen-4-ol	1177	0.672	0.907	1.825	0.353
24	p-cymen-8-ol	1183	nd	nd	nd	0.354
25	italidione (MW 170)	1191	nd	nd	nd	0.929
26	α-terpineol	1191	0.973	1.412	1.054	0.751
27	myrtenol	1195	nd	nd	nd	0.333
28	verbenone	1205	nd	nd	nd	0.846
29	cis-geraniol (nerol)	1228	1.289	2.766	0.407	nd
30	cis-citral (neral)	1240	nd	0.504	nd	nd
31	neryl acetate	1364	19.872	23.222	32.297	18.272
32	α- copaene	1376	nd	nd	2.406	nd
33	isoitalicene	1395	1.098	nd	1.457	0.615
34	italicene	1403	nd	1.334	nd	1.266
35	cis-α-bergamotene	1415	2.001	0.959	0.169	nd
36	β-caryophyllene	1419	2.612	2.251	1.995	nd
37	trans α-bergamotene	1435	2.135	0.941	0.226	nd
38	italidione (MW 210)	1449	8.412	10.968	5.726	8.472
39	geranyl propionate	1475	4.185	7.912	5.445	5.402
40	2-octen-1-ol, 3,7-dimethyl-, isobutyrate, (Z)-	/	nd	nd	nd	6.053
41	γ-himachalene	1477	1.975	nd	nd	nd
42	α-curcumene	1483	nd	7.421	10.035	9.970
43	β-selinene	1486	14.811	nd	nd	nd
44	italidione (MW 224)	1493	2.096	2.496	1.801	3.466
45	α-selinene	1494	4.594	nd	nd	nd
46	γ-cadinene	1513	nd	nd	0.561	nd
47	calamenene	1523	nd	nd	0.342	nd
48	(6S,10R-dimethylbicyclo [4.4.0]decan-1-en3-one)	1529	0.722	2.242	1.726	7.737

49	α-calacorene	1542	nd	nd	0.234	nd
50	diepicedrene-1-oxide	1551	nd	nd	nd	0.249
51	2-octen-1-ol, 3,7-dimethyl-, isobutyrate, (Z)-	/	nd	nd	nd	1.812
52	caryophyllene oxide	1583	nd	nd	3.790	6.457
53	italidione (MW 238)	1583	0.857	0.877	nd	nd
54	trans nuciferol	/	nd	nd	nd	0.840
55	copaborneol	/	nd	nd	1.847	0.673
56	rosifoliol	1600	3.665	11.294	5.416	1.557
57	selin-6-en-4α-ol	1636	0.489	nd	nd	nd
58	10,10-dimethyl-2,6-dimethylenebicyclo[7.2.0]undecan-5β-ol	1644	nd	nd	nd	0.568
59	β-eudesmol	1649	0.165	0.528	nd	2.076
60	eudesm-7(11)-en-4-ol	1692	0.939	nd	nd	nd
61	ent-germacra-4(15),5,10(14)-trien-1β-ol	1695		nd	nd	0.892
Σ (%)			100.0	99.8	100.0	99.6

RI - Retention indice; nd - not detected

In our study, the composition of volatile constituents varied notably among the four treatments (alfalfa mulch, straw mulch, perennial mulch, and control), with clear patterns emerging when compounds were considered in retention index order. Early-eluting monoterpene hydrocarbons, such as α-pinene, β-pinene, β-myrcene, and limonene, were abundant in all samples, with their highest levels observed in alfalfa mulch, where α-pinene (11.44%) and limonene (6.07%) were particularly dominant. Oxygenated monoterpenes, including eucalyptol, linalool, terpinen-4-ol, and α-terpineol, were especially well represented in straw mulch, with linalool (1.97%) and α-terpineol (1.41%) exceeding levels found in other treatments.

Mid- to late-retention compounds were dominated by sesquiterpenes and esters, with neryl acetate being a key differentiating component, peaking in perennial mulch at 32.30%. Italidiones of various molecular weights (MW 170, 210, 224, and 238) were detected in all treatments, sometimes at substantial concentrations, such as italidione MW 210 in straw mulch (10.97%), suggesting their presence is less dependent on mulch type and may be influenced by inherent plant material or soil transformation processes. The control sample generally contained lower proportions of monoterpenes but exhibited a distinct profile of oxygenated sesquiterpenes and late-eluting volatiles, including 2-octen-1-ol, diepicedrene-1-oxide, and β-eudesmol, some of which were absent in mulched treatments.

Overall, mulching type strongly influenced the composition and relative abundance of volatile compounds. Alfalfa mulch was characterised by monoterpene richness, perennial mulch by high neryl acetate content, and control samples by unique oxygenated sesquiterpenes. These compositional differences likely reflect the combined effects of plant residue chemistry, microbial activity, and environmental conditions under each treatment.

These differences are likely attributable to changes in soil temperature induced by mulch application. Previous research (Ning *et al.*, 2021) found that soil under black mulch, such as perennial mulch, was warmer than unmulched controls (Palada *et al.*, 2000). In our study, the warmer soil beneath black mulch likely stimulated plant growth and neryl acetate synthesis, while the cooler soil in the unmulched treatment may have inhibited its production. This interpretation is supported by two key factors. First, temperature is one of the most critical environmental variables affecting the growth and yield of medicinal plants (Gavrić *et al.*, 2023). Secondly, average temperatures at the research location are below those in the Mediterranean region, where immortelle is most grown.

Although our research has provided insight into the impact of mulching on inflorescence yield, EO content and bioactive components of immortelle, it has some limitations. A limitation of this research is that the research was conducted on a one-year-old crop, and data on the maximum potential yield of immortelle could not be obtained. Therefore, this research should be continued, and knowledge should be gained about the impact of mulch on yield and quality in terms of economic yields. Despite these limitations, our research showed certain advantages that can be applied in practice. Namely, the tested mulches had a positive effect on the yield and quality. Therefore, its more intensive application in sustainable agriculture could be a cheap alternative to environmentally unfavorable methods of weed control, while simultaneously preserving moisture in the soil and improving the yield and quality of the cultivated species.

Conclusions

The experimental findings demonstrated that mulching significantly influences inflorescence yield, EO content, and bioactive compound composition in immortelle. Alfalfa mulch resulted in a notably higher yield of inflorescences compared to other mulch types (perennial mulch, straw, and control). Furthermore, the choice of mulch had a substantial impact on the quality of the EO. More precisely, the use of perennial mulch increased the content of neryl acetate in the EO. These results suggest that increased mulch application could serve as a cost-effective strategy to enhance both the yield and quality of immortelle. However, given the relatively low overall yield observed in this study, further research is recommended in subsequent growing seasons once the plants reach full maturity.

Authors' Contributions

Conceptualization: AC, KHV, TG; Data curation: KVH, TG; Formal analysis: AC, KVH; Funding acquisition: TG; Investigation: AC, KVH, TG; Methodology: TG; Project administration: TG; Resources: AC, TG; Software: KVH, TG; Supervision: TG; Validation: KHV, TG; Visualization: TG; Roles/Writing - original draft: AC, KHV, TG; and Writing - review & editing: AC, KHV, TG.

All authors read and approved the final manuscript.

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Conflict of Interests

The authors declare that there are no conflicts of interest related to this article.

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