

## Chemical Composition and *in vitro* Antioxidant Activity of *Mutellina purpurea* Thell. Flowers Essential Oil

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**Abstract:** *Mutellina purpurea* L. belongs to Apiaceae family and is known as Alpin lovage considered as a forage for animals nutrition because of the presence of sufficient concentration of minerals like calcium and potassium. The chemical composition of the essential oil obtained by hydrodistillation in a Clevenger apparatus and aroma of inflorescences of *Mutellina purpurea* (Poir.) Thell. was analyzed by GC/MS and GC-FID. Sabinene (19.2%),  $\alpha$ -pinene (12.6%), (*E*)-sesquisabinene hydrate (9.0%), myrcene (7.8%), (*Z*)-sesquisabinene hydrate (7.5%) and  $\alpha$ -bisabolol (6.7%) dominated in the essential oil obtained by hydrodistillation. Static headspace technique gave sabinene (23.0%), *p*-cresol (17.4%),  $\alpha$ -pinene (17.0%) and myrcene (10.9%) as a major constituents. Antioxidant activity of the oil was evaluated by ABTS assay.

**Keywords:** *Mutellina purpurea*; essential oil; hydrodistillation; static headspace technique.  
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### 1. Plant Source

The *Mutellina purpurea* (Poir.) Thell (Apiaceae) flowers were collected in the Botanical Garden of the Medical University in Lublin in June 2010. The voucher specimen has been deposited at the Herbarium of the Department of Pharmacognosy with Medicinal Plant Unit, Medical University in Lublin (ES032011M).

### 2. Previous Studies

Concluding, hitherto the literature is lacking the chemical composition of *M. purpurea* flowers essential oil. In the present work the *M. purpurea* flowers essential oil and aroma are described and compared.

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### 3. Present Study

#### 3.1. Essential oil and aroma composition

From the fresh plant material 16.8 mL/kg (dry weight) of the yellowish oil with a specific odour, in the Clevenger-type apparatus, was obtained. The list of detected compounds with their relative percentages and retention indices are given in Table 1 in order of their elution. The aroma-composition obtained by static headspace technique (SHS) is also given in Table 1. Sixty-six compounds in the essential oil obtained in Clevenger-type apparatus (C-EO) were counted whereas static headspace technique yielded fifty-one volatile compounds (Table 1). Fifty-seven compounds were identified by GC/MS in the hydrodistilled oil which accounted for around 86% of the total oil composition. The headspace-GC analysis allowed to identify forty-seven compounds (92%). This oil was dominated by monoterpenoids. The main compounds of the C-EO were sabinene (19.2%),  $\alpha$ -pinene (12.6%), myrcene (7.8%), (*E*)-sesquisabinene hydrate (9.0%), (*Z*)-sesquisabinene hydrate (7.5%), and  $\alpha$ -bisabolol (6.7%). The main compounds of the aroma were sabinene (23.0%), *p*-cresol (17.4%),  $\alpha$ -pinene (17.0%) and myrcene (10.9%). Hydrodistillation (HD) yielded bicyclic sesquiterpenes as sesquisabinene hydrate (*Z*)- and (*E*)- (7.5-9%) and monocyclic sesquiterpen  $\alpha$ -bisabolol (6.7%) whereas these compounds in the headspace-GC analysis constitute less than 0.15%. The aroma contained more monoterpene hydrocarbons such as  $\alpha$ -pinene, *p*-cymene, myrcene, *p*-cresol,  $\gamma$ -terpinene and *trans*- $\beta$ -ocimene. The content of oxygenated monoterpene sabinene was different in the aroma and in the C-EO (19.2 and 23.0%, respectively). The amount of  $\alpha$ -pinene,  $\beta$ -pinene, and  $\alpha$ -phellanderene was higher in the aroma than in the C-EO. These results are partially against Okoh et al. (2010), where the hydrodistilled oil contained more monoterpene hydrocarbons such as  $\alpha$ -pinene, camphene,  $\beta$ -pinene, myrcene,  $\alpha$ -phellanderene, 1,8-cineole, *trans*- $\beta$ -ocimene and  $\gamma$ -terpinene [1]. Hydrodistillation in Clevenger-type apparatus and static headspace technique yielded the same dominant components, however apart from this C-EO contained 12 other compounds which percentage were higher than 1% in the sum, the aroma contained 9 of such compounds respectively.

Hydrodistillation of *Prangos pabularia* Lindl. (Apiaceae) yielded 72.9% of high-boiling compounds (sesquiterpenoids, phenylpropanoids), while microdistillation method gave only 53.7% of these compounds [2]. Basically, the same trend was observed in previous studies on volatiles from Apiaceae plants: *Prangos turcica* A. Duran, M. Sađirođlu & H. Duman [3], *Angelica sylvestris* L. var. *sylvestris* [4], *Rhabdosciadium oligocarpum* (Post ex Boiss.) Hedge & Lamond and *Rhabdosciadium microcalycinum* Hand.-Mazz. [3], where different extraction methods were compared. It was shown that HD was particularly effective in the isolation of the high-boiling compounds. Despite the long time of hydrodistillation process may cause the artifacts [5] the infinite distillation time affects the higher content of many compounds. SHS is the simplest, the most sensitive, and the quickest method to analyze the fragrant components of the plants, which are less decomposed by temperature.

**Table 1.** The composition of *M. purpurea* flowers essential oil obtained in Clevenger-type apparatus (C-EO) and by means of static headspace technique (SHS).

No.	Compound	RI <sub>exp</sub> <sup>a</sup>	C-EO		SHS	
			Relative amount (%) <sup>b</sup>	SD (%)	Relative amount (%) <sup>b</sup>	SD (%)
1	$\alpha$ -thujene	931	0.2	-	0.4	-
2	<b><math>\alpha</math>-pinene</b>	938	<b>12.6</b>	0.1	<b>17.0</b>	4.9
3	camphene	953	1.3	-	1.8	0.3
4	<b>sabinene</b>	974	<b>19.2</b>	0.2	<b>23.0</b>	0.9
5	$\beta$ -pinene	979	1.5	-	3.8	0.1
6	<b>myrcene</b>	988	<b>7.8</b>	-	<b>10.9</b>	2.9
7	$\alpha$ -phellanderene	1008	nd	-	0.2	-
8	$\alpha$ -terpinene	1017	nd	-	0.8	0.1
9	<i>p</i> -cymene	1023	1.8	-	2.2	0.2
10	<b>limonene</b>	1027	<b>3.6</b>	-	<b>2.3</b>	0.1

11	$\beta$ -phellandrene	1029	0.5	-	1.7	0.2
12	( <i>Z</i> )- $\beta$ -ocimene	1033	1.0	-	1.9	1.0
13	( <i>E</i> )- $\beta$ -ocimene	1043	2.2	-	3.3	2.3
14	$\gamma$ -terpinene	1055	0.5	-	2.9	0.5
15	( <i>Z</i> )-sabinene hydrate	1067	0.2	-	nd	-
16	<b><i>p</i>-cresol</b>	1070	0.3	-	<b>17.4</b>	2.0
17	terpinolene	1081	0.1	-	0.4	0.2
18	linalool	1095	0.2	-	0.1	-
19	( <i>E</i> )-sabinene hydrate	1097	0.2	-	t	-
20	( <i>Z</i> )- <i>p</i> -menth-2-en-1-ol	1121	0.1	-	0.1	-
21	( <i>E</i> )- <i>p</i> -menth-2-en-1-ol	1139	nd	-	0.2	-
22	unidentified compound-I	1168	nd	-	4.8	0.2
23	terpinen-4-ol	1182	0.7	0.1	1.7	0.1
24	$\alpha$ -terpineol	1194	nd	-	0.3	0.1
25	( <i>Z</i> )-sabinene hydrate acetate	1220	nd	-	t	-
26	carvacrol methyl ether	1242	0.1	-	t	-
27	lavandulyl acetate	1287	0.2	-	0.1	-
28	bornyl acetate	1290	0.3	-	0.2	-
29	unidentified compound-II	1386	0.1	-	nd	-
30	$\beta$ -elemene	1393	1.8	-	0.2	-
31	methyl eugenol	1404	0.1	-	0.1	-
32	2,5-dimethoxy- <i>p</i> -cymene	1418	0.3	-	t	-
33	( <i>E</i> )- $\beta$ -caryophyllene	1427	2.4	0.1	0.3	-
34	$\gamma$ -elemene	1436	0.1	-	nd	-
35	( <i>E</i> )- $\alpha$ -bergamotene	1440	t	-	t	-
36	( <i>Z</i> )- $\beta$ -farnesene	1460	0.7	-	0.2	0.1
37	$\alpha$ -humulene	1467	0.2	-	t	-
38	germacrene D	1470	2.4	0.2	0.2	0.1
39	dehydro-sesquicineole	1478	1.9	0.1	0.1	-
40	$\beta$ -acoradiene	1488	0.1	-	0.1	0.1
41	$\gamma$ -curcumene	1491	nd	-	0.1	0.1
42	bicyclogermacrene	1510	1.6	0.1	0.2	0.1
43	$\alpha$ -chamigrene	1512	nd	-	t	-
44	$\beta$ -bisabolene	1515	t	-	nd	-
45	$\alpha$ -selinene	1519	t	-	t	-
46	germacrene A	1521	0.8	-	nd	-
47	sesquicineole	1525	0.2	-	t	-
48	$\delta$ -amorphene	1531	0.1	-	nd	-
49	unidentified compound-III	1537	nd	-	t	-
50	unidentified compound-IV	1550	t	-	nd	-
51	$\beta$ -sesquiphellandrene	1554	0.1	-	nd	-
52	unidentified compound-V	1559	0.1	-	nd	-
53	<b>(<i>Z</i>)-sesquisabinene hydrate</b>	1564	<b>7.5</b>	-	t	-
54	germacrene B	1571	1.2	-	nd	-
55	unidentified compound-VI	1576	nd	-	0.2	-
56	spathulenol	1588	1.4	-	nd	-
57	caryophyllene oxide	1593	0.3	-	t	-
58	<b>(<i>E</i>)-sesquisabinene hydrate</b>	1598	<b>9.0</b>	-	t	-
59	viridiflorol	1605	0.1	-	nd	-
60	$\beta$ -atlantol	1624	0.2	-	nd	-
61	unidentified compound-VII	1630	0.1	-	nd	-
62	1- <i>epi</i> -cubenol	1639	0.2	-	nd	-
63	$\alpha$ -acorenol	1647	1.1	-	nd	-
64	<i>epi</i> - $\alpha$ -muurolol	1658	0.2	-	nd	-
65	unidentified compound-VIII	1662	0.1	-	nd	-
66	$\beta$ -acorenol	1666	0.3	-	nd	-
67	$\alpha$ -cadinol	1671	0.6	-	nd	-
68	<i>neo</i> -intermedeol	1675	0.2	-	nd	-
69	$\beta$ -bisabolol	1685	1.5	-	t	-
70	bulnesol	1691	0.2	-	t	-
71	<b><math>\alpha</math>-bisabolol</b>	1703	<b>6.7</b>	-	0.1	0.1
72	( <i>Z,Z</i> )-farnesol	1712	1.0	-	nd	-
73	( <i>Z</i> )- $\alpha$ -bisabolene epoxide	1726	0.1	-	nd	-
74	unidentified compound-IX	1750	0.1	-	nd	-
75	unidentified compound-X	1770	nd	-	0.7	0.1

76	unidentified compound-XI	1827	0.3	-	nd	-
77	unidentified compound-XII	2141	t	-	nd	-
<b>Total detected</b>			<b>100.0</b>		<b>100.0</b>	

<sup>a</sup> RI<sub>exp</sub>: experimental Retention Indices with respect to a series of n-alkanes. <sup>b</sup> Average values (peak area relative to total peak area) from three replicate sample analyses. The predominant compounds are indicated in bold. t – traces, for less than 0.05 %; nd – not detected; SD - standard deviation; "-" SD < 0.05

### 3.2. The antioxidant assay

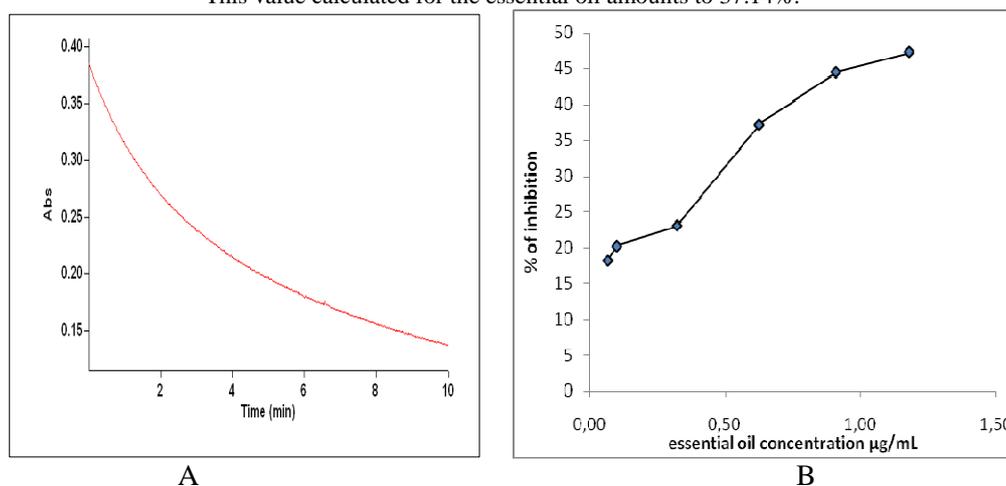
The dilutions of essential oil in n-pentane were used because the inhibition of ABTS•+ caused by pure essential oil was too high to draw the relationship. The EC<sub>50</sub> value was calculated as a dilution giving 50 percent inhibition of ABTS radical cation solution. The EC<sub>50</sub> value (Effective Concentration) for *M. purpurea* flowers essential oil was calculated as 1.18 µg/mL. Figure 1A presents the kinetic reaction between the ABTS radical cation and the essential oil dilution 1.18 µg/mL. The relationship between the percent of inhibition and the dilutions of essential oil is presented in Figure 1B. The Q value presents the ratio between the amount of neutralized radical and the primary amount of radical and was expressed in %.

$$Q = \frac{100\%(A_0 - A_p)}{A_0}$$

A<sub>0</sub> – the absorbance of pure radical (without the antioxidant)

A<sub>p</sub> – the absorbance of radical mixed with antioxidant [6].

This value calculated for the essential oil amounts to 37.14%.



**Figure 1.** A - The kinetic reaction between the ABTS radical cation and the essential oil dilution 1.18 µg/mL; B - The relationship between the percent of inhibition and the dilutions of essential oil.

Comparing EC<sub>50</sub> value obtained for *M. purpurea* flowers essential oil with other essential oils from Apiaceae family, the *M. purpurea* essential oil was more active. The essential oils from *Ligusticum chuanxiong* Hort. and *Cnidium officinale* Makino exhibited an EC<sub>50</sub> value of 1.58 mg/mL for both oils in the ABTS test [7]. In general, the antioxidative effectiveness of essential oil depends on the content of phenolic compounds and the reaction activity of the phenol towards the chain-carrying peroxy radicals and on the stability of the phenoxyl radical formed in the reaction [8]. In the investigated essential oil the oxygenated mono- and sesquiterpenes constituted 20% of all compounds. Among them *p*-cresol, linalool, methyl eugenol, (*Z*)-*p*-menth-2-en-1-ol, terpinen-4-ol, β-atlantol, 1-*epi*-cubenol, α-acorenol, *epi*-α-muurolool, β-acorenol, α-cadinol, *neo*-intermedeol, β-bisabolol, bulnesol, α-bisabolol, (*Z,Z*)-farnesol, carvacrol methyl ether, α-bisabolol and β-bisabolol were detected in the highest content. α-Bisabolol accounted for 6.7% and β-bisabolol 1.5% of the sum of all detected compounds. It is known that phenols are efficient antioxidants and many of them have been tested. For instance thymol and carvacrol are compounds with demonstrated antioxidant activity [9, 10, 11].

Summing up, the chemical composition of the essential oil from *M. purpurea* flowers makes it an antioxidant with Q value 37.14% for a 0.625 µg/mL concentration.

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### Supporting Information

Supporting Information accompanies this paper on <http://www.acgpubs.org/RNP>

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