

CHEMICAL COMPOSITION AND FUNGICIDAL ACTIVITY OF THE ESSENTIAL OIL OF *Laserpitium garganicum* FROM ITALY

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Laserpitium garganicum subsp. *garganicum* (Ten.) Bertol. (= *Laserpitium siler* L. subsp. *garganicum* (Ten.) Arcangeli) is a perennial herb belonging to the Apiaceae family. The distribution is limited to the southern area of the Balkan peninsula and Italy [1]. In Italy this plant is found in the central Apennines, Sicily and Sardinia [2]. This plant is described as a subspecies of *L. siler* or a species of *Laserpitium* in the Flora Europaea [1] and the Flora d'Italia [2] respectively. A few studies have reported the biologically active components isolated from *L. siler*, mainly sesquiterpene lactones [3–8], and one refers to sesquiterpene lactones from the roots of *L. garganicum* [9]. The essential oil composition of *L. siler* was also reported [10–13], but to the best of our knowledge, this is the first report on the GC/MS determination of the essential oil composition of the *L. garganicum* subsp. *garganicum* (Ten.) Bertol. Since the biological activity of this plant has not been studied, we focused our investigation on the antifungal activities of this essential oil against some phytopathogens and opportunistic human fungi.

Fifty-six compounds were identified in *L. garganicum* essential oil, representing 92.3% of the total oil [14–19]. Table 1 shows the list of components identified and their percentages and retention indices. Compounds are listed in order of their elution from an HP-5. The most abundant compounds were myrcene (15.7%), β -phellandrene (14.4%), sabinene (9.7%), and γ -muurolene (7.8%).

Monoterpene hydrocarbons made up 47.0% of the total oil; myrcene (15.7%) was the most abundant compound. Significant amounts of sesquiterpene hydrocarbons (25.0%) were found, with γ -muurolene (7.8%) being the main component. The oxygenated sesquiterpene fraction made up 9.8% of the total oil, with spathulenol (4.0%) having the highest content. The oxygenated monoterpenes (7.7%) also contributed with a similar content to the essential oil. This fraction was dominated by terpinen-4-ol (4.3%). Esters and ketones represented 1.8% and 0.6% of the total oil, respectively. The oil of *L. garganicum* differs markedly from the fruit oil of *L. siler* from southern France [13]; the later is characterized by perillaldehyde (75.0%) and limonene (22.0%), compounds that were not found in *L. garganicum* oil. The oil from fruits of *L. siler* analyzed by Motl [10] contained perillaldehyde (89.5%) and limonene (10.5%).

According to Adcock and Betts [11] the lack of perillaldehyde and limonene in *L. garganicum* is reason to consider these plants as a species of *Laserpitium* instead of a subspecies of *L. siler* despite the morphological similarities. Based on chemical composition, the plants analyzed in the present work were more similar to the other *Laserpitium* species reported by Adcock and Betts [11] (*L. prutenicum* L., *L. hispidum* Bieb., *L. glaucum* L., *L. halleri* Crantz, *L. krapfii* Crantz, *L. archangelica* Wulfen, *L. latifolium* L., and *L. gallicum* L.) than to *L. siler*.

The antimycotic activity of essential oils depends on their chemical composition and may play a fundamental role in the host/pathogen relationship. Systemic fungal infections are important problems in medicine. Infections caused by fungal species are common in immunocompromised patients and result in significant treatment costs and mortality.

Table 2 shows the antimicrobial activity of the essential oil of *L. garganicum*.

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TABLE 1. Chemical Composition of the *L. garganicum* Essential Oil

Component	KI*	Percentage	Component	KI*	Percentage
α -Pinene	937	5.7	(<i>E</i>)-Caryophyllene	1406	1.7
Sabinene	978	9.7	β -Copaene	1417	0.4
Myrcene	997	15.7	γ -Elemene	1426	0.7
α -Terpinene	1021	0.2	Aromadendrene	1429	0.1
β -Phellandrene	1031	14.4	<i>cis</i> -Muurola-3.5-diene	1433	0.3
<i>trans</i> - β -Ocimene	1053	0.4	α -Humulene	1441	0.6
γ -Terpinene	1060	0.4	<i>allo</i> -Aromadendrene	1454	0.1
<i>cis</i> -Sabinene hydrate	1071	0.1	γ -Muurolene	1474	7.8
Terpinolene	1088	0.5	β -Selinene	1476	0.7
<i>trans</i> -Sabinene hydrate	1099	0.1	<i>cis</i> - β -Guaiene	1479	0.1
Myrcenol	1124	0.4	Bicyclogermacrene	1488	4.1
<i>trans</i> -Pinocarveol	1140	0.2	<i>trans</i> - β -Guaiene	1490	1.0
<i>trans</i> - <i>p</i> -Menth-2-en-1-ol	1145	0.3	α -Muurolene	1492	0.1
<i>trans</i> -Verbenol	1149	0.2	γ -Cadinene	1502	0.2
Pinocarvone	1163	0.1	δ -Cadinene	1515	1.1
Borneol	1166	0.2	Zonarene	1528	0.2
Terpinen-4-ol	1178	4.3	Elemol	1545	2.5
α -Terpineol	1189	1.8	Germacrene B	1555	0.2
Myrtenol	1194	0.2	(<i>E</i>)-Nerolidol	1564	0.1
<i>cis</i> -Piperitol	1207	0.2	Spathulenol	1571	4.0
Bornyl formate	1222	1.4	Viridiflorol	1580	0.4
<i>trans</i> -Carveol	1271	0.2	Guaiol	1622	0.6
Bornyl acetate	1282	0.4	<i>epi</i> - α -Muurolol	1634	0.4
Bicycloelemene	1329	1.3	β -Eudesmol	1639	0.6
α -Copaene	1366	0.3	α -Eudesmol	1643	0.4
β -Bourbonene	1374	0.9	α -Cadinol	1647	0.8
β -Cubebene	1381	0.1	Neophytadiene	1834	0.2
β -Elemene	1385	3.0	Phytol	2110	0.2

*Retention indices relative to *n*-alkane series on an HP-5 column.

TABLE 2. Antimicrobial Activity of the Essential Oil of *L. garganicum*

Microorganism	% Inhibition*			
	0.125 μ L/mL**	0.250 μ L/mL**	0.5 μ L/mL**	1 μ L/mL**
<i>A. niger</i>	21 \pm 7	31 \pm 6	32 \pm 4	28 \pm 4
<i>A. terreus</i>	n.i.	14 \pm 5	17 \pm 5	22 \pm 6
<i>C. globosum</i>	n.i.	22 \pm 3	22 \pm 4	20 \pm 4
<i>P. chrisogenum</i>	n.i.	10 \pm 4	15 \pm 5	47 \pm 5
<i>P. pinophilum</i>	23 \pm 6	28 \pm 5	34 \pm 5	54 \pm 4
<i>T. viride</i>	13 \pm 4	33 \pm 3	42 \pm 4	67 \pm 2

*The data are the mean of triplicate values \pm SD.

**Essential oil content (μ L/mL cultured medium).

n.i.: no inhibition.

The minimal oil concentration (0.0625 μ L/mL) did not have any effect on the tested microorganism. At the maxima concentration tested, the oil exhibited good antifungal activities against *T. viride* (67% \pm 2), *P. pinophilum* (54% \pm 4), and *P. chrisogenum* (47% \pm 5). Other interesting antimycotic activities were observed against *A. niger* (28% \pm 4), *A. terreus*

(22% ± 6), and *C. globosum* (20% ± 4). *A. niger* is sensitive to the oil at a concentration of 0.125 µL/mL and above, but the inhibition remains practically constant for all the tested concentrations. *A. terreus* began to respond to the oil at a 0.25 µL/mL concentration, and the inhibition increased almost linearly with oil concentration. *C. globosum* shows a moderate response that was independent of the oil concentrations tested. *P. chrisogenum* had a logarithmic increase of the inhibition from 0.25 µL/mL to 1 µL/mL. *T. viride* showed a rapid increase from 0.125 µL/mL to 0.25 µL/mL followed by an inhibition increase that was quite linear until a concentration of 67% ± 2; this microorganism was the most sensitive to the inhibition activity of the tested oil.

From the observed results, it can be concluded that the oil of *L. garganicum* has a strong antifungal activity. This activity could be due to the presence of the oxygenated components.

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