





## The inhibitory activity of *Inula viscosa* essential oil against Solanaceae fungal strains: A case of eggplant (*Solanum melongena*)

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### ABSTRACT

The study, conducted with the aim of protecting market garden crops, focused on the medicinal plant *Inula viscosa*, whose essential oil was evaluated for its fungicidal efficacy against phytopathogenic fungi affecting *Solanum melongena* L. (eggplant). The essential oil yield was relatively high, approximately 1.53%. The major constituents of *Inula viscosa* essential oil were germacrene D (12.7%),  $\delta$ -cadinene (9.6%), and  $\alpha$ -cadinol (8.5%). The essential oil demonstrated notable effectiveness, with mycelial growth inhibition rates for the tested fungal strains – *Epicoccum* sp., *Geotricum* sp., *Aspergillus niger*, and *Trichoderma* sp. – ranging from 2.35% to 100%, depending on the concentration used. A comparative study of the inhibitory efficacy of *Inula viscosa* essential oil and the fungicide Vapcotop revealed the superior activity of the essential oil against *Epicoccum* sp., *Geotricum* sp., and *Aspergillus niger*. However, *Trichoderma* sp. was more sensitive to Vapcotop than to the essential oil. These promising results support the recommendation of *Inula viscosa* essential oil as a natural alternative to synthetic fungicides, contributing to the protection of Solanaceae crops while preserving both environmental and human health.

**Keywords:** biopesticide, *Inula viscosa*, *Solanum melongena*, fungal strains, inhibition.

### INTRODUCTION

A member of the Solanaceae family, eggplant (*Solanum melongena*) is a tropical vegetable and the seventh most widely consumed vegetable in the world (FAO, 1989). It plays an important economic role in Asia, Africa and the subtropic regions, and is also grown in temperate regions, such as the Mediterranean and the southern United States (Djouadi and Lanez, 2012). Vegetable crops, particularly eggplants, are often subjected to biotic and abiotic stresses factors, causing disturbances in their

metabolism. Biotic stresses are due to the plant's interaction with other organisms such as fungi, bacteria, viruses, etc. These pathogens, by infecting plants and particularly market garden crops, will affect their growth as well as yield, and may cause them to perish (Hassena, 2009). Conventional cultural and chemical control methods against these pathogens have their limits. In fact, synthetic plant protection products, long regarded as effective in improving yields, are now being questioned due to their demonstrated toxicity to human and animal health., as well as their ecotoxic effects on the

environment. It is therefore becoming imperative to restrict their use by implementing alternative strategies capable of limiting the recourse to synthetic chemical substances. This is why new so called “biological” control strategies aim to induce defense mechanisms in plants, using natural products such as medicinal plants. *Inula viscosa*, as a biological control agent due to its medicinal nature, is an annual herbaceous plant, slimy and glandular, with a strong odor, belonging to the Asteraceae family, with antimicrobial and antifungal activities. It has been widely used to extend the shelf life of food and in traditional medicine (Adam et al., 1998) as an antipyretic, antiseptic and anti-inflammatory agent (Djerroumi and Nacef, 2004).

The leaves of *Inula viscosa* secrete a mixture of resins throughout the life of the leaves (Cafarchia and al., 2002); these exudates consist of several aglycone flavonoides, as well as numerous terpenoids (Cafarchia et al., 2001). They show significant allelopathic activity, as well as an inhibitory effect on phytopathogenic microorganisms (Stavrianakou et al., 2006). In order to evaluate the inhibitory effect of *Inula viscosa* essential oil on four fungal strains isolated from the studied eggplants, namely *Epicoccum* sp., *Geotricum* sp., *Trichoderma* sp. and *Aspergillus niger*, which have been identified as phytopathogenic organisms of Solanacea, the authors focused on the extraction and analysis of the constituent elements of the essential oil and its application in order to test its efficacy. Subsequently, the chemical pesticide Vapcotop, for agricultural use, was used as a comparative treatment and antifungal control.

Although several studies have reported the antifungal properties of essential oils, limited information is available concerning the specific activity of *Inula viscosa* essential oil against major phytopathogenic fungi of eggplant. This knowledge gap hinders the identification of eco-friendly alternatives to synthetic fungicides, which are increasingly required in sustainable agriculture. Therefore, the central question arises:

- How, at the present time, in a development perspective and an environment in a perpetual climatic and edaphic changes, would the exploitation of *Inula viscosa* essential oil, allow the protection of a market-garden plantation suffering attacks from phytopathogenic fungi?
- How do natural essential oils compare with the Vapcotop pesticide?

It was hypothesized that *Inula viscosa* essential oil exhibit significant antifungal activity capable of inhibiting the growth of major phytopathogenic fungi of eggplant.

Further it was assumed that their richness in secondary metabolites may account for this biological potential and provide a basis for the development of sustainable alternatives to chemical fungicides. It is in the context of novel technologies derived from plant biology, that the aim of the conducted study was to highlight the phytochemical composition and inhibitory potential of *Inula viscosa* essential oil against eggplant pathogens, with a view to assessing its suitability as a biocontrol agent for the protection of market garden plants.

## MATERIALS AND METHODS

### Plant material

The plant studied was *Inula viscosa*, the aerial parts, stems and leaves were collected in April in the Commune of Beni-Snous, in the Wilaya of Tlemcen, north-west Algeria, and left to dry at room temperature, protected from light and humidity. After drying, the samples were stored in bags until extraction. The specimen of the voucher was deposited at the herbarium of the University of Tlemcen under the number IMtAP.325.02.2022.

### Extraction method

Essential oils were extracted by hydrodistillation according to the Meyer-Warnod method (Meyer-Warnod, 1984), using a Clevenger-type apparatus compliant with the European Pharmacopoeia (Européenne-Pharmacopée, 1997). Dried plant material (400 g) was placed in a flask with 600 mL of distilled water and heated for 3 to 4 hours. The essential oil was carried by the steam, and the vapors were condensed in a condenser, yielding a distillate with a thin layer of essential oil on the surface. After phase separation, the oil was recovered from the top of the separatory tube using a Pasteur pipette. It was then stored in an opaque vial at 4 °C in the dark.

### Yield calculation

The essential oil yield (EOY) is calculated as the ratio of the weight of the essential oil obtained

(Wo, in grams) to the weight of the dried plant material used (Wp, in grams) (Afnor, 1986). It is expressed as a percentage (%) using the following formula:

$$EOY = \left( \frac{W_o}{W_p} \right) \times 100 \quad (1)$$

### Analysis by gas chromatography

The essential oil was analyzed using gas chromatography (GC) and gas chromatography–mass spectrometry (GC/MS). GC analysis was performed on a Perkin Elmer AutoSystem GC equipped with a Flame Ionization Detector (FID), a single injector, and two fused-silica capillary columns (60 m × 0.22 mm i.d., 0.25 µm film thickness) featuring different stationary phases: Rtx-1 (polydimethylsiloxane) and Rtx-Wax (polyethylene glycol). The temperature program ranged from 60 °C to 220 °C, increasing at a rate of 2 °C per minute. For GC/MS analysis, a Perkin Elmer TurboMass detector was used under the same chromatographic conditions and with the same columns as those employed in the GC analysis. The ion source temperature was set at 150 °C, with an electron ionization energy of 70 eV. Mass spectra were recorded in the range of 35–350 Da, using a split ratio of 1:80. The components were identified by: (i) comparing their gas chromatographic retention indices (GC-RIs) on both non-polar and polar columns calculated by linear interpolation relative to a series of n-alkanes with those of authentic standards or published literature data (Jennings and Shibamoto, 1980; N.I.S.T., 2008); and (ii) matching their mass spectra with those from commercial databases (Mc Lafferty and Stauffer, 1988; N.I.S.T., 1999) as well as with spectra from an in-house spectral library.

### Antifungal activity

#### Isolation of fungal strains

Segments of eggplant, taken randomly from the study station at the Technical Institute for Vegetable and Industrial Crops (ITCMI) in Hassi Bounif (Wilaya of Oran, Algeria), were placed under aseptic conditions seeding on Petri dishes containing isolation medium Potato Dextrose Agar (PDA: potato 200 g, Dextrose 20 g and Agar 15 g/L diluted at 25 °C) then incubated at 28 °C for seven days. After growth of several fungal strains, four fungi (the most dominants) were isolated: *Epicoccum*

sp., *Geotricum* sp., *Trichoderma* sp. and *Aspergillus niger*, then maintained by transplanting onto PDA medium in order to purify them. Isolated and purified fungi were stored at 4 °C.

#### Inoculum preparation

Sixteen Petri dishes were prepared, four for each fungal strain: one tin containing only the fungus and the culture medium (PDA), the other three containing the culture medium, the fungus and the disc of Whatman paper of 6 mm soaked in *Inula viscosa* essential oil. The simplest technique was used, that of direct seeding: after running a thin layer of 4 mm of the gel medium (20 mL) previously liquefied with PDA, the fungi *Epicoccum* sp., *Geotricum* sp., *Trichoderma* sp. and *Aspergillus niger* were inoculated into the 16 dishes using a platinum loop previously sterilized.

#### "In-vitro" test of antifungal activity

The method employed was that of direct confrontation: in the center of each box, a sterile disk of Whatman paper with 6 mm diameter was placed, using a pair of sterile forceps, and then added 10 µL of the different concentrations of *Inula viscosa* essential oil, diluted in Dimethyl Sulfoxide (DMSO), tested: 0.3 µL/mL, 0.5 µL/mL, 0.6 µL/mL, 0.75 µL/mL and 0.83 µL/mL.

For each concentration, three replicates were performed. All Petri dishes were incubated at 28° ± 4 °C for 7 days. The results were read by measuring the diameter of the zone of inhibition of fungal strain growth, around the discs soaked with the different concentrations of oils tested.

The percentage inhibition of mycelial growth (Ic) was calculated using the following formula:

$$Ic = \frac{Di \times 100}{Dc} \quad (2)$$

where: *Di* – diameter of mycelial growth inhibition in the vicinity of the oil-bearing disk;  
*Dc* – diameter of mycelial growth in the control box.

### Approach to statistical processing and evaluation of results

The methods applied in this work are based on analysis of variance (ANOVA) to assess the statistical significance of treatment effects, along with the calculation of lethal concentrations (LC<sub>10</sub>, LC<sub>50</sub>, LC<sub>90</sub>) using probit regression, in order to estimate the essential oil doses required to

achieve fungal growth inhibition rates ranging from 10% to 90% in. Statistical analyses were performed with SPSS software, with significance level set at ( $P < 0.05$ ).

### Positive control tests

In addition to the essential oil studied, positive control tests were carried out to compare its level of antifungal activity with that of Vapcotop, a chemical fungicide widely used by ITCMI in the Wilaya of Oran. This fungicide was applied at the recommended dosage to control the various phytopathogenic fungal strains, with reference to its technical data sheet. The fungicide solution was prepared by diluting 0.7 g/L of Vapcotop powder in sterile distilled water. We used the same quantity of essential oil (10  $\mu$ L) to compare the inhibitory effect of the fungicide against our essential oil, at its lowest concentration (0.3  $\mu$ L/L). They were tested on the three strains most present on the studied eggplants: *Geotricum* sp., *Aspergillus niger* and *Trichoderma* sp.

## RESULTS

### Essential oil yield

Essential oil yield (EOY) was calculated as a function of dried vegetable matter. In the study of *Inula viscosa*, it was 1.53%, rather high compared with other plants tested (Silano and Delbó, 2008; Aprotosoiaie and al. 2010).

### Chemical composition of essential oil

The essential oil of *Inula viscosa* was analyzed using gas chromatography–mass spectrometry (GC-MS), enabling the identification of 65 compounds, its constituent 90.2% of total compounds of the essential oil (Table 1).

The analyzed essential oil exhibits in Table 1 a composition rich in sesquiterpenes, particularly both hydrocarbon (30.7%) and oxygenated sesquiterpenes (58.5%), which represent the major fraction of its volatile profile. The predominant component was germacrene D (12.3%), followed by  $\delta$ -cadinene (9.6%) and  $\alpha$ -cadinol (8.5%). These compounds contribute to the characteristic aroma of the oil, often described as woody and spicy. Other notable constituents include tau-cadinol (7.5%), shyobunol (6.3%),  $\beta$ -oplopenone

(4.5%) and viridiflorol (4.1%). The oil also contains appreciable amounts of caryophyllene oxide (2.5%), ar-curcumen-15-al (2.2%), germacrene D-4-ol (2.1%),  $\alpha$ -muurolene (1.2%) and  $\gamma$ -cadinene (1.5%). The composition is further enriched by various other sesquiterpenes in lower proportions, such as ledol, nerolidol isomers, and several cadinane-type derivatives. The high content of oxygenated compounds contributes to the complex aromatic profile of oils, combining woody, earthy and slightly balsamic notes.

### The antifungal activity of the essential oil of *Inula viscosa* against the different strains (*Geotricum* sp., *Epicoccum* sp., *Trichoderma* sp. and *Aspergillus niger*)

#### Estimation of lethal concentration values

The lethal concentrations were calculated to establish the concentrations of essential oil of *Inula viscosa* that determine the inhibition for the different trains (*Geotricum* sp., *Epicoccum* sp., *Trichoderma* sp. and *Aspergillus niger*) (Table 2).

The previous table leads to the conclusion that the lethal effects of essential oil of *Inula viscosa* on the different trains (*Geotricum* sp., *Epicoccum* sp., *Trichoderma* sp. and *Aspergillus niger*) has a very significant effect: *Epicoccum* sp. demonstrated the highest sensitivity, with a  $LC_{50}$  of 0.49  $\mu$ L/mL and  $LC_{90}$  of 0.78  $\mu$ L/mL while *Aspergillus niger* showed a  $LC_{50}$  of 22.53  $\mu$ L/mL and a greatest resistance  $LC_{90}$  of 781,74  $\mu$ L/mL.

### Essential oil activity on molds

The antifungal activity of *Inula viscosa* essential oil was tested against various phytopathogenic strains isolated and purified from our eggplants: *Epicoccum* sp., *Geotricum* sp., *Trichoderma* sp. and *Aspergillus niger*. The results obtained are shown in (Figure 1). Figure 1 shows the mycelial growth inhibition rates at various concentrations of *Inula viscosa* essential oil, demonstrating significant but contrasting inhibition rates against the four fungal strains, thus demonstrating its antifungal power. These results were compared with negative controls for each strain for which a mycelial mat was observed. In fact, the antifungal activity of *Inula viscosa* essential oil against *Epicoccum* sp. showed remarkable inhibition rates (Figure 2), ranging from 20.11%, 21.52%, 69.88% and 92.58%



**Table 1.** Chemical composition of *Inula viscosa* essential oil by GC-MS

No <sup>a</sup>	Compounds	Lit /RIa <sup>b</sup>	RIa <sup>c</sup>	RIp <sup>d</sup>	EO (%)	Identification <sup>e</sup>
1	Hexanal	773	770	1055	0.1	RI, MS
2	Z-hex-3-en-1-ol	834	831	1380	0.1	RI, MS
3	β-citronella	938	940	1031	tr	RI, MS
4	6-methyl-hept-5-en-2-one	961	963	1337	tr	RI, MS
5	1,8-Dihydro cinéole	979	979	1180	0.2	RI, MS
6	Octanal	981	982	1169	0.2	RI, MS
7	Nonal	1082	1083	1394	0.2	RI, MS
8	α-Terpineol	1179	1175	1688	0.1	RI, MS
9	Theaspirane 1	1293	1292	1480	0.1	RI, MS
10	Cis-3-hexenyl tiglate	1305	1304	1641	0.1	RI, MS
11	Theaspirane 2	1308	1307	1517	0.1	RI, MS
12	α-cubebene	1348	1350	1452	0.1	RI, MS
13	α-ylangene	1375	1372	1476	0.2	RI, MS
14	α-Copaene	1379	1379	1488	0.1	RI, MS
15	β-borbonene	1383	1385	1515	0.1	RI, MS
16	Cyprene	1402	1406	1525	0.3	RI, MS
17	Cis-α-bergamotene	1409	1411	1562	0.1	RI, MS
18	α-ghurjunene	1413	1410	1524	0.8	RI, MS
19	Aristolene	1420	1416	1560	tr	RI, MS
20	E-β-caryophyllene	1424	1418	1524	0.4	RI, MS
21	β-copane	1426	1431	1581	0.1	RI, MS
22	β-ghurjunene	1439	1439	1591	0.2	RI, MS
23	E-β-farnesene	1448	1448	1660	0.3	RI, MS
24	Nomadendrene	1452	1450	1581	0.1	RI, MS
25	α-humulene	1456	1451	1611	0.3	RI, MS
26	Allo-aromadendrene	1462	1463	1476	0.3	RI, MS
27	α-Curcumene	1471	1468	1465	0.4	RI, MS
28	γ-murolene	1471	1471	1681	0.4	RI, MS
29	Germacrene D	1480	1480	1704	12.3	RI, MS
30	Zingebrene	1486	1489	1717	0.2	RI, MS
31	α-murolene	1496	1994	1719	1.2	RI, MS
32	E,E-α-farnesene	1498	1500	1740	0.2	RI, MS
33	γ-cadinene	1507	1507	1752	1.5	RI, MS
34	Trans-calamenene	1512	1511	1816	0.4	RI, MS
35	δ-cadinene	1516	1516	1752	9.6	RI, MS
36	α-cadinene	1535	1530	1740	1.1	RI, MS
37	α-calacorene	1531	1531	1895	0.6	RI, MS
38	β-calacorene	1548	1547	1936	0.3	RI, MS
39	E-nerolidol	1546	1551	2037	1.4	RI, MS
40	Epi-globulol	1558	1559	2010	0.4	RI, MS
41	Caryophyllene oxyde	1576	1571	1980	2.5	RI, MS
42	Z-nerolidol	1518	1521	1987	0.3	RI, MS
43	Germacrene D-4-ol	1573	1566	2020	2.1	RI, MS
44	Globulol	1580	1576	2067	0.2	RI, MS
45	Humulene epoxyde	1592	1601	2044	0.6	RI, MS
46	β-oplophenone	1593	1590	2017	4.5	RI, MS
47	Viridiflorol	1591	1591	2089	4.1	RI, MS

48	Zingiberenol	1598	1599	2019	0.5	RI, MS
49	1,10-diepi-cubenol	1608	1608	2025	0.5	RI, MS
50	Aromadendrene epoxyde	1615	1617	2002	1.1	RI, MS
51	Eudesma-4(15) -en-6-one	1616	1614	2039	0.6	RI, MS
52	Cadin-4-en-7-ol	1627	1627	2096	1.4	RI, MS
53	Trans-murolol	1630	1630	2134	0.6	RI, MS
54	Tau-cadinol	1632	1632	2169	7.5	RI, MS
55	$\beta$ -eudesmol	1638	1644	2234	tr	RI, MS
56	$\alpha$ -carinol	1644	1645	2231	tr	RI, MS
57	$\alpha$ -cadinol	1645	1642	2231	8.5	RI, MS
58	Z,Z-farnesol	1648	1653	2167	1.8	RI, MS
59	$\alpha$ -eudesmol	1653	1649	2220	1.4	RI, MS
60	$\beta$ -bisabolol	1656	1653	2140	1.3	RI, MS
61	Ar-curcumen-15-al	1681	1675	2164	2.2	RI, MS
62	Eudesma-7,11-en-4- $\alpha$ -ol	1683	1683	2300	0.4	RI, MS
63	Shyobunol	1687	1688	2218	6.3	RI, MS
64	Ledol	1696	1693	2050	1.7	RI, MS
65	E-acetate de nerolidol	1732	1738	2269	3.1	RI, MS
66	$\beta$ -acoradienol	1797	1792	2221	2.7	RI, MS
Total					90.2	

**Note:** <sup>a</sup> Order of elution is given on apolar column (Rtx-1). <sup>b</sup> Retention indices of literature on the apolar column (RIa) reported from König et al, 2001. <sup>c</sup> Retention indices on the apolar Rtx-1 column (RIa). <sup>d</sup> Retention indices on the polar Rtx-Wax column (RIp). <sup>e</sup> RI: Retention Indices; MS: Mass Spectra in electronic impact mode. EO: Essential oil.

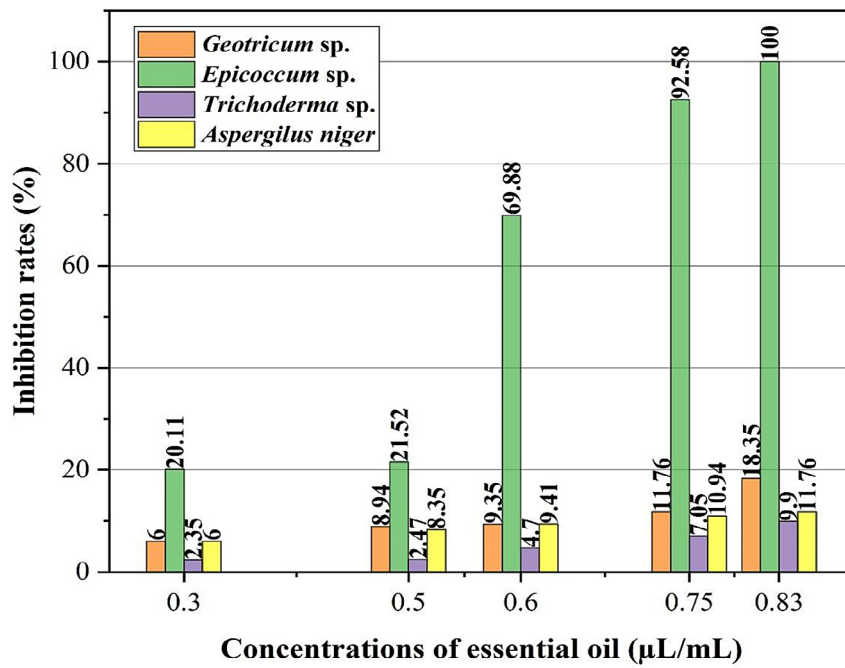
**Table 2.** Estimated values of lethal concentration (LC) after essential oil of *Inula viscosa* treatment of the different trains (*Geotricum* sp., *Epicocum* sp., *Trichoderma* sp. and *Aspergillus niger*)

LC %	<i>Inula viscosa</i> $\mu\text{L/mL}$			
	<i>Geotricum</i> sp.	<i>Epicocum</i> sp.	<i>Trichoderma</i> sp.	<i>Aspergillus niger</i>
10	0.54	0.30	0.93	0.65
20	1.15	0.36	1.66	2.19
30	1.98	0.40	2.51	5.28
40	3.15	0.45	3.58	11.17
50	4.86	0.49	4.99	22.53
60	7.51	0.54	6.94	45.42
70	11.95	0.59	9.89	96.18
80	20.59	0.67	14.96	231.38
90	43.77	0.78	26.57	781.74

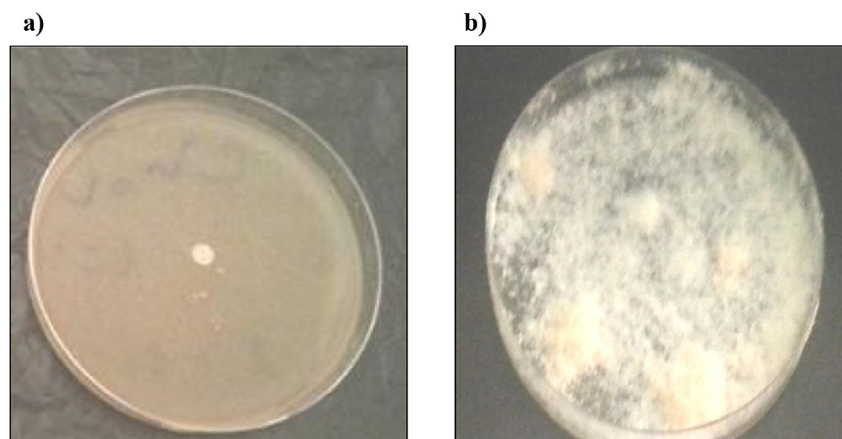
at concentrations of 0.3  $\mu\text{L/mL}$ , 0.5  $\mu\text{L/mL}$ , 0.6  $\mu\text{L/mL}$  respectively, reaching 100% at 0.83  $\mu\text{L/mL}$ .

In addition, the antifungal activity of *Inula viscosa* essential oil was equally inhibitory against *Geotricum* sp. and *Aspergillus niger*, with inhibition rates of 6%, 8.94%, 9.35%, 11.76% and 18.35% for *Geotricum* sp. at concentrations of 0.3  $\mu\text{L/mL}$ , 0.5  $\mu\text{L/mL}$ , 0.6  $\mu\text{L/mL}$ , 0.75  $\mu\text{L/mL}$  and 0.83  $\mu\text{L/mL}$  respectively

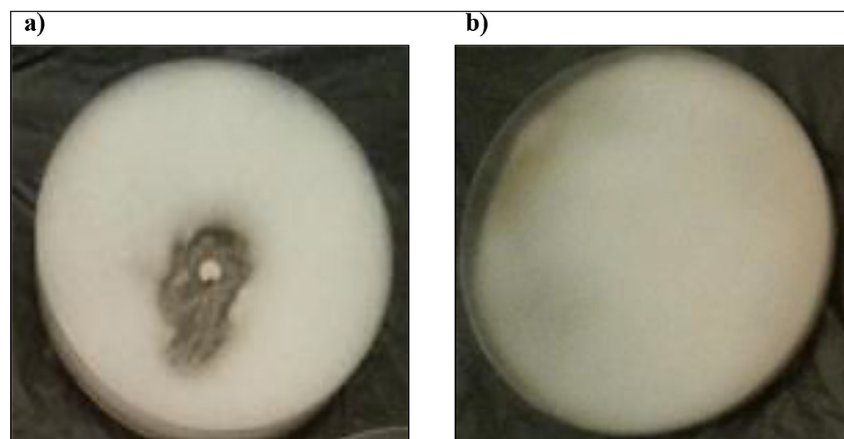
(Figure 3). For *Aspergillus niger* (Figure 4), the inhibition rates ranged from 6% to 11.75% at the same concentrations as above. A weak inhibitory activity was found against *Trichoderma* sp. with minimal inhibition percentages of around 9.9% at the highest concentration of 0.83  $\mu\text{L/mL}$  (Figure 5). For all strains tested, fungal growth inhibition rates were proportional to essential oil concentrations.



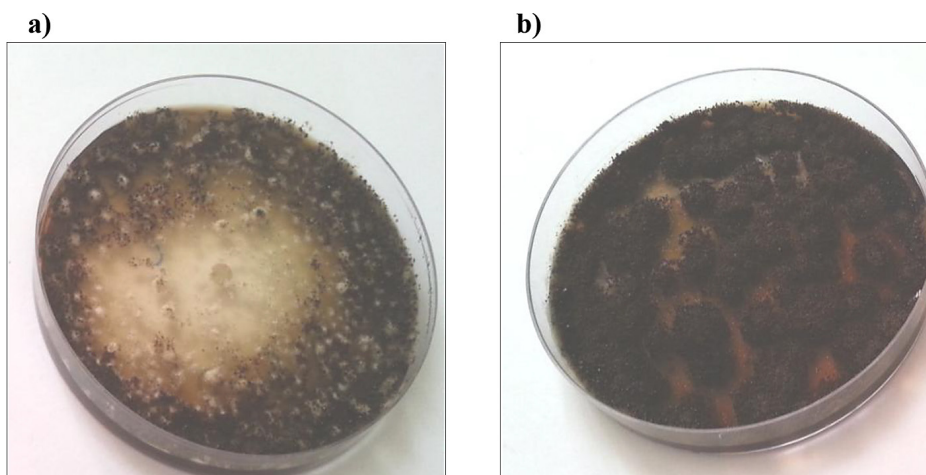
**Figure 1.** The mycelial growth inhibition rates at various concentrations of *Inula viscosa* essential oil



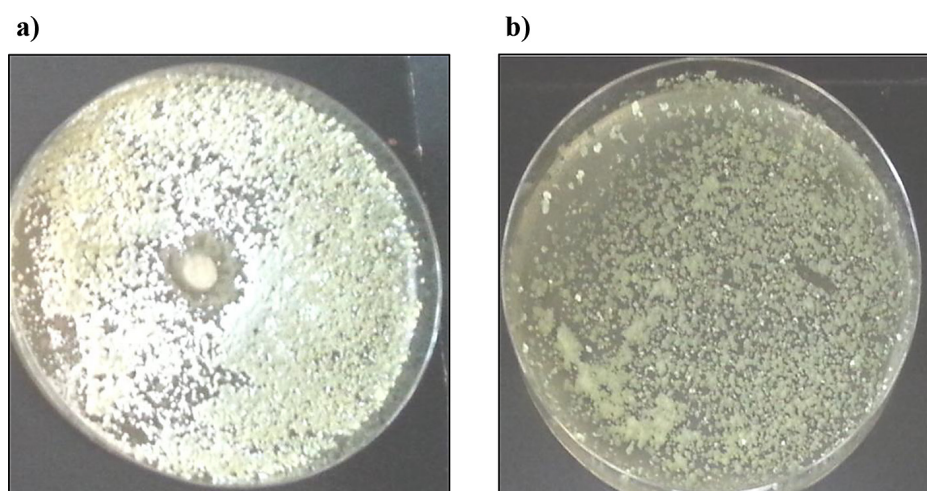
**Figure 2.** Photos showing the inhibitory power of *Inula viscosa* essential oil against *Epicoccum* sp. at concentration  $C = 0.83 \mu\text{L/mL}$  (a) and control (b)



**Figure 3.** Photos showing the inhibitory activity of *Inula viscosa* essential oil on *Geotricum* sp. at concentration  $C = 0.83 \mu\text{L/mL}$  (a) and the control (b)



**Figure 4.** The inhibitory effect of *Inula viscosa* essential oil on *Aspergillus niger* at concentration  $C = 0.83 \mu\text{L/mL}$  (a) and control (b)



**Figure 5.** The inhibitory effect of *Inula viscosa* essential oil on *Trichoderma* sp. at concentration  $C = 0.83 \mu\text{L/mL}$  (a) and the control (b)

### Comparison of the antifungal activity of essential oils with that of Vapcotop

In order to compare the efficacy of the essential oil at the lowest concentration ( $0.3 \mu\text{L/L}$ ) with that of Vapcotop ( $0.7 \text{ g/L}$ ) against the three molds *Geotricum* sp., *Aspergillus niger* and *Trichoderma* sp., the same volume ( $10 \mu\text{L}$ ) was used for both products. The results are shown in the following photos: (Figures 6, 7 and 8). We found that the minimal concentration used ( $0.3 \mu\text{L/mL}$ ) of *Inula viscosa* essential oil was more effective than Vapcotop, since its activity was nil at the higher concentration. Its activity was nil at the recommended concentration of  $0.7 \text{ g/L}$  (Figures 6 and 7). However, it was found that the use of the fungicide Vapcotop gave a more significant result

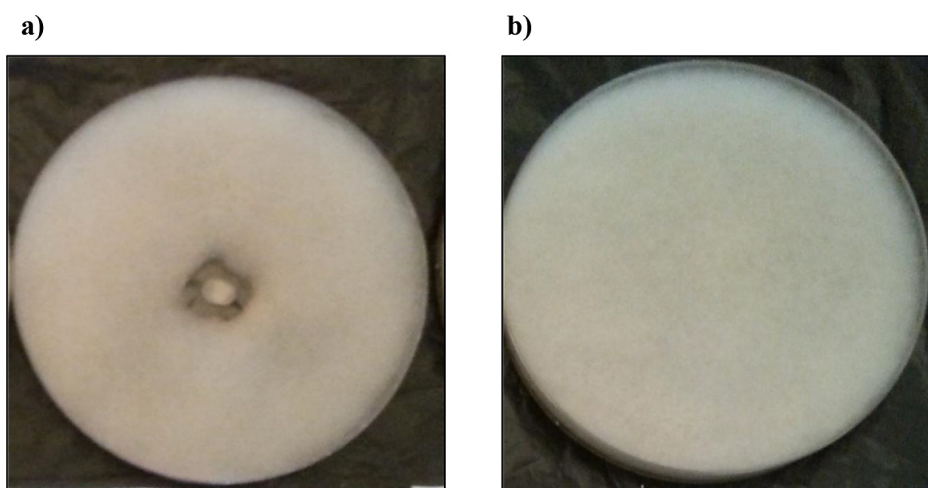
against *Trichoderma* sp. than the essential oil of *Inula viscosa* (Figure 8).

### DISCUSSION

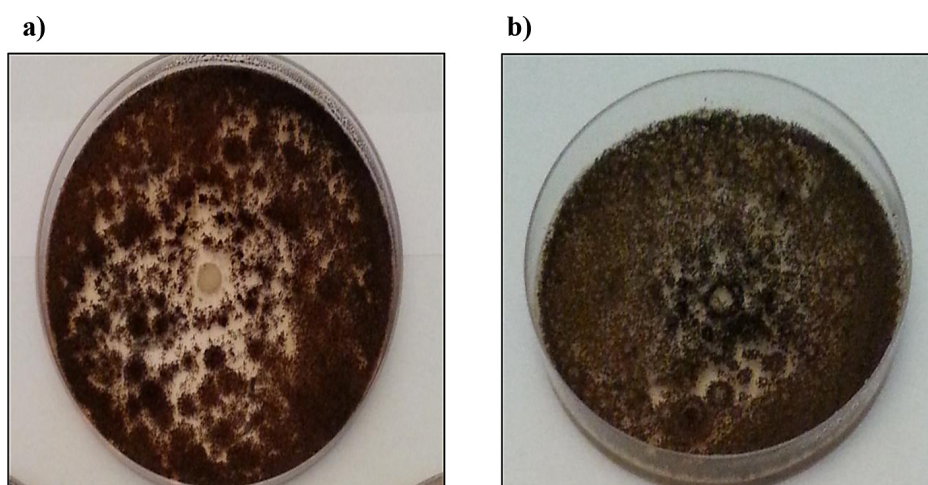
The conducted study showed that the yield of essential oil from *Inula viscosa* was high compared with other studies, where it ranges from  $0.03\%$  to  $0.07\%$  (Blanc et al., 2006),  $0.09\%$  (Ms-sillou et al.; 2022),  $0.148\%$  (Haoui et al., 2015),  $0.15\%$  (Ainseba et al., 2023) and  $0.30\%$  (Miguel et al., 2008).

This variation may be ascribed to several factors, including the plant's geographical origin, prevailing environmental and climatic conditions, as well as the drying process, period and

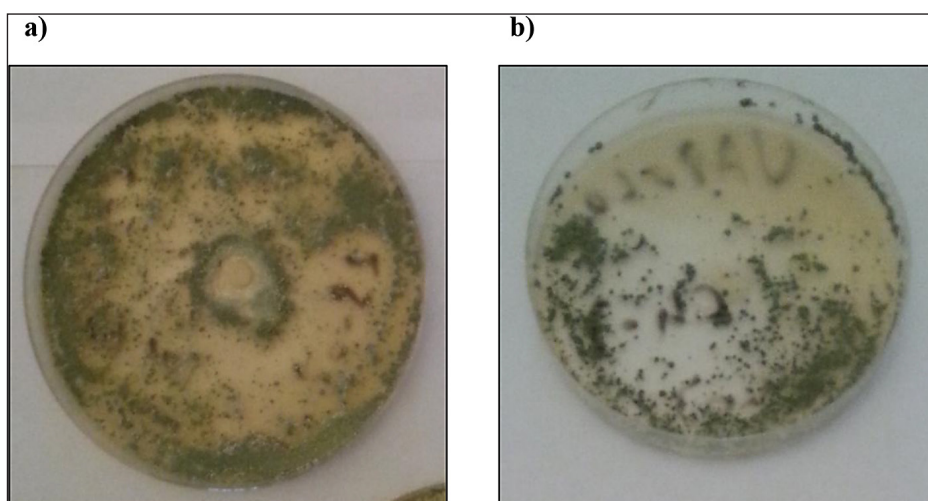




**Figure 6.** The antifungal activity of *Inula viscosa* essential oil at a concentration of 0.3  $\mu\text{L/mL}$  on *Geotricum* sp. (a) compared with that of the Vapcotop fungicide (0.7 g/L) (b)



**Figure 7.** The antifungal activity of *Inula viscosa* essential oil at a concentration of 0.3  $\mu\text{L/mL}$  with the fungal strain *Aspergillus niger* (a), compared with that of the Vapcotop fungicide (0.7 g/L) (b)



**Figure 8.** The antifungal activity of *Inula viscosa* essential oil at a concentration of 0.3  $\mu\text{L/mL}$  on *Trichoderma* sp. (a), compared with that of the fungicide Vapcotop (b)

harvesting location (Silano and Delbó, 2008; Aprotosoaie et al., 2010; Olle and Bender, 2010). The age of the plant material and the extraction technique, as well as the part of the plant studied, can also influence the yield (Garnero, 1977). An in depth look at the microbiological field in parallel with the medicinal field allow observing a scientific reality in which the use of essential oils is considered an “effective” alternative for inhibiting fungal growth. This relatively widespread use reflects the antifungal efficacy of essential oils in general, and of *Inula viscosa* in particular.

The obtained results show that this essential oil has good antimicrobial activity against the strains isolated from the eggplants used in this study. Statistical analysis of the inhibition rates, combined with the estimation of lethal concentrations allowed for a quantitative evaluation of the oil efficacy of in a dose-dependent manner.

The relatively low  $LC_{50}$  values indicate a strong fungistatic or even fungicidal potential, depending on the concentration applied. This suggests that certain bioactive constituents present predominantly in *Inula viscosa* essential oil such as terpene: Viridiflorol,  $\beta$ -acoradiénol and  $\alpha$ -curcumen-15-al, sesquiterpenoid like cadinol ( $\alpha$ -cadinol, Tau-cadinol), Nerolidyl acetate, sesquiterpene (Germacrene D) or bicyclic sesquiterpene ( $\delta$ -cadinene), sesquiterpenoids monocyclic compounds like shyobunol, and epoxide (Humulene epoxide) may play a key role in inhibiting mycelial growth.

Indeed, studies on the chemical composition of *Inula viscosa* essential oil show a complex structure with 80% of different components, mainly sesquiterpene; terpenes, are the majority constituents (Kheyar and al., 2014).

Previous works (Benchohra et al., 2011; Haoui et al., 2015) reported that the antimicrobial effect is attributed particularly to terpenes inducing structural perturbations of the membrane, thereby increasing its permeability and resulting in the efflux of cytoplasmic constituents (Juven et al., 1994; Ultee et al., 1999; Knowles et al., 2005; López-Malo et al., 2005; El Ajjour et al., 2008).

However, it should be remembered that it is the overall composition that has the effect. In other words, mycelial inhibition is not the result of the effect of a single component, but of the synergy and interaction of the various constituents; the resulting action of the various probable chemical interactions gives rise to a significant oil response.

Compared with the agricultural antifungal Vapcotop, the low-concentration essential oil of *Inula viscosa* was more effective against *Aspergillus niger* and *Geotricum* sp., although inhibition rates were not very high. On the other hand, for *Trichoderma* sp. a change in color from white to yellowish was clearly noted, probably the result of an interruption in conidial growth. Conidial growth is usually marked by a natural change from yellow to green. Furthermore, the percentage inhibition of Vapcotop against *Trichoderma* sp. was higher, which would be due to its chemical composition: L e-methyl-thiophanate (Vapcotop 70, Pelt) belonging to the Benzimidazole family (D.P.P.T.C., 2015). This bicyclic compound is a combination of benzene and imidazole. Benzimidazole and its derivatives are used as fungicides because they inhibit the action of some micro-organisms (Bettiche, 2017).

## CONCLUSIONS

In a context of generalization of biological control in the face of industrial chemical control; the results of the conducted study have shown the effectiveness of the process defined in the conducted work for antifungal control, the use of essential oil as a biopesticide represents an interesting alternative for the protection of crops against fungi. The natural product tested, in this case *Inula viscosa* essential oil, proved to be suitable and advantageous for the protection of market garden plants, the latter being the main link in the biotope, therefore its use is recommended to protect crops, the environment and, consequently, consumers.

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