



Biological control with essential oil of *Foeniculum vulgare* Mill

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Abstract

The present study was aimed to investigate, the chemical composition of the essential oil extracted by hydrodistillation from Fennel (*Foeniculum vulgare* Mill.) harvested in South-Est, Algeria, and to evaluate its antifungal and allelopathic activities.

Gas chromatography-mass spectrometry GC-MS analysis of *F. vulgare* essential oil showed the presence of 11 components, alpha-phellandrene (29.44%) was found as the main component. The essential oil was tested on the seeds of 2 crops (*Chenopodium quinoa* Willd.) Variety Q102 and (*Triticum durum* Desf.) variety vitron, The final germination percentages and the seedling shoot and root lengths were significantly reduced by the *F. vulgare* essential oil as compared to the control. Furthermore, the antifungal study on the fungi of *Aspergillus niger* and *Aspergillus flavus* reported significant results. The effectiveness of the oil showed that it has a fungicidal effect against the two strains. These findings suggest that the *F. vulgare* essential oil can be used as a source of environmental-friendly in biological control.

Keywords: Allelopathie, Antifungal activity, *Foeniculum vulgare* Mill., Essential oil.

Foeniculum Vulgare Mill'in Uçucu Yağı İle Biyolojik Kontrol

Öz

Bu çalışmanın amacı, Cezayir'in Güney-Doğu bölgesinde hasat edilen Rezene'den (*Foeniculum vulgare* Mill.) hidrodistilasyon yoluyla elde edilen uçucu yağın kimyasal bileşimini araştırmak ve antifungal ve allelopatik aktivitelerini değerlendirmektir.

Gaz kromatografisi-kütle spektrometrisi *F. vulgare* uçucu yağının GC-MS analizi, 11 bileşenin varlığını göstermiş ve alfa-phellandrene (%29.44) ana bileşen olarak bulunmuştur. Uçucu yağ, 2 mahsulün (*Chenopodium quinoa* Willd.) Çeşit Q102 ve (*Triticum durum* Desf.) çeşidinin vitron tohumları üzerinde test edilmiştir. Kontrol amaçlı karşılaştırıldığında *F. vulgare* esansiyel yağı ile nihai çimlenme yüzdeleri ve fide sürgün ve kök uzunlukları önemli ölçüde azaltılmıştır. Ayrıca, *Aspergillus niger* ve *Aspergillus flavus* mantarları üzerinde yapılan antifungal çalışma önemli sonuçlar bildirmiştir. Yağın etkinliği, iki suşa karşı mantar öldürücü bir etkiye sahip olduğunu göstermiştir. Bu bulgular, *F. vulgare* esansiyel yağının biyolojik mücadelede çevre dostu bir kaynak olarak kullanılabileceğini düşündürmektedir.

Anahtar Kelimeler: Allelopathie, Antifungal aktivite, *Foeniculum vulgare* Mill., Uçucu yağ.

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1. Introduction

The use of herbicides has a harmful effect on the environment and public health. Accordingly it is important to alternatives for controlling weeds. An alternative suggested by the author, to control weeds, is the development and application of control measures with plant products (Alves et al., 2014). Many plants synthesize toxic substances for defense against other plants and microorganisms including viruses, bacteria, and fungi. (Liman et al., 2012, Kucukboyac et al. 2011) Allelopathy is a biological phenomenon by which an organism produces one or more biochemicals that influence the growth, survival, and reproduction of other organisms (El Ayeb et al., 2014).

The lamiaceae family is one of the most widely used families as a global source of spices and extracts with strong antimicrobial and antioxidant properties. (Goudjil et al., 2020) Fennel is a member of the family Apiaceae. It is classified into two sub species: subsp. Capillaceum which includes three varieties (azoricum Miller, dulce Miller (sweet fennel) and vulgare), Subsp. Piperitum (pepper fennel)., (Muckensturm et al., 1997; Marotti et al., 1994; Piccaglia and Marotti, 2001).

The plant and its essential oil have been extensively used as carminative, digestive, galactagogue and diuretic and to treat respiratory and gastrointestinal disorders (Farukh et al., 2017). It is also used as a constituent in cosmetic and pharmaceutical products, the plant and its essential oil have been extensively used as carminative, digestive, galactagogue and diuretic and to treat respiratory and gastrointestinal disorders (Mimica-Dukic et al., 2003). It is also used as a constituent in cosmetic and pharmaceutical.

2. Material and Method

2.1. Plant material

The aerial parts of Wild fennel *F. vulgare* harvested in February 2021 in the region of Elmarouthia (South-East Algeria) then dried in the shade at an ambient temperature during 10 days then hydrodistilled using a Clevenger-type apparatus for 3 h to give an essential oil yield of 0.89%.

2.1. Gas chromatography-mass spectrometry analysis

The analysis of *F. vulgare* EO was performed at the scientific and technical research center in physico-chemical analyzes (CRAPC).. The apparatus used is a gas chromatograph of the (TQ 8040 NX) type coupled with a mass spectrometer. quadrupole ionization voltage of 70 ev. The column that is used is an HP-5MS; 5% Phenyl Methyl Siloxane with a length of 30 m and an internal diameter of 0.25 mm. The wire thickness being 0.25 mm. The operating conditions are:

- The temperature of the injector (split mode 1:20): 250 °C
- Temperature programming: from 70 °C to 280 °C at a rate of 4 °C/min.

- The vector gas used is helium with a flow rate of 0.79 ml/min. The temperatures of the quadrupole source are fixed, respectively, at 250 °C and 280 °C. Linear retention indices (RI) for all compounds were determined using n-alkanes as standards. Identification of individual compounds was performed by matching their mass spectral fragmentation patterns with corresponding data available (Wiley 275 library (6th edition)).

2.2. Allelopathic activity

Each concentration was prepared with diluted *F. vulgare* EO in DMSO then added sterile distilled water to give final concentrations of : 0.5 ; 1 ; 2 : 3 %. All germination tests are performed in sterile glass Petri dishes with a diameter of 90 mm and a height of 18 mm. Standard filter discs equal to the diameter of the plates are placed in Petri dishes. After we put 25 quinoa seeds in a Petri dish and 10 wheat seeds in another one. Then pour a few milliliters (enough to cover only the seeds) of each concentration in each Petri dish and for each concentration, we kept three replicates. Germination is indicated by taking out the roots from the seed layer at least 2 mm long. The germination rate corresponds to the maximum percentage of germinated seeds compared to the total seeds sown, it estimated by the following formula (Alves et al., 2014):

$$GP\% = (NI / NT) \times 100$$

Where **GP**: germination percentage (%).

NI: The number of seeds germinated.

NT: The total number of seeds sown.

The inhibitory or stimulatory percent was calculated using the following equation given by Chung et al., 2001:

$$\text{inhibition (-) / stimulation (+) \%} = [(\text{extract} - \text{control}) / \text{control}] \times 100,$$

where 'extract' is the parameter measured in the presence of the *F. vulgare* EO and 'control' is the parameter measured in the presence of distilled water.

2.3. Anti-fungal activity

The antifungal activity was to examine the chemical composition of the essential oil extracted from the *F. Vulgare* against two species: *Aspergillus niger* and *Aspergillus flavus* prepared in the biology laboratory of the University of Ouargla.

The method used according to goudjil et al., with slight modifications. Is the direct contact method where the 04 concentrations in which concentration is obtained by adding of essential oil to 30 ml lukewarm Potatoes Dextrose Agar PDA in a bottle plus 0.5% DMSO. After shaking the flasks, the medium is poured into plastic dishes 90 mm in diameter. The inoculation is done under the Biological safety cabinets by depositing in the center of the dish a disc of the mycelial about 6mm in diameter. Petrie dish containing 15ml of PDA medium plus DMSO and the other only PDA, both without oil essential are inoculated to serve as a control. The Petri dishes (controls and tests) are incubated at 25 ± 2 ° C respectively for 7 days. All tests, are restarted three times (Goudjil et al., 2020).

Table 1. Chemical composition of Algerian *F. Vulgare* EO

Copounds	Retention time (min)	Area (%)
β-Pinene	4.17	0.6
alpha.-phellandrene	4.39	29.44
o-Cymene	4.53	10.11
D-Limonene	4.58	19.48
Sabinene	4.61	/
α-Terpinolene	5.11	0.22
Dill ether	6.10	21.52
(E)-Pinocarveol	6.27	0.34
trans-Sabinol	6.65	0.14
(-)-Carvone	6.74	9.76
Durenol	7.36	2.18

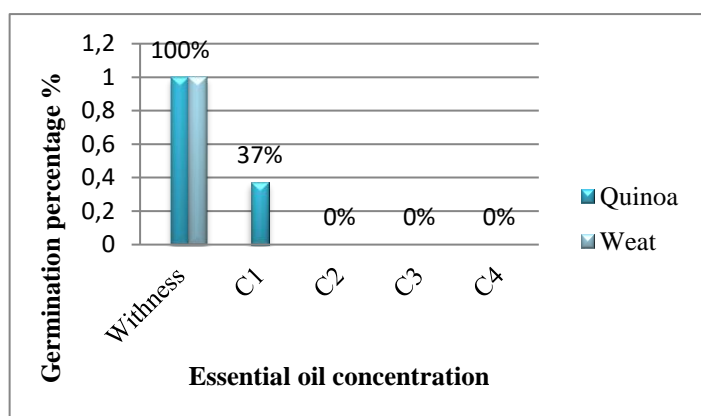


Figure 1. Germination percentage of Wheat and quinoa seeds treated with *F.vulgare* oil

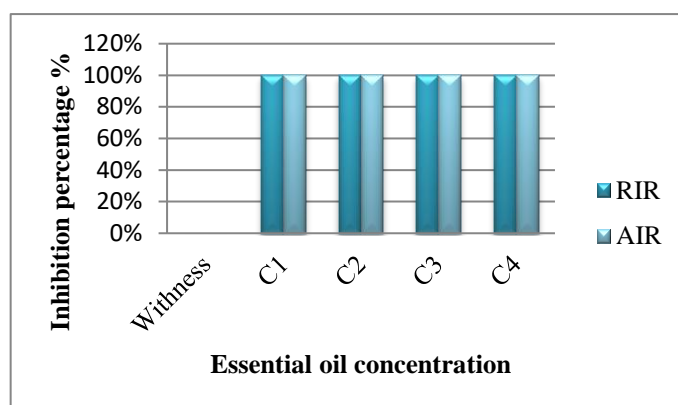


Figure 2. Inhibition of RIR and AIR recorded for Wheat seeds treated with *F.vulgare* oil.

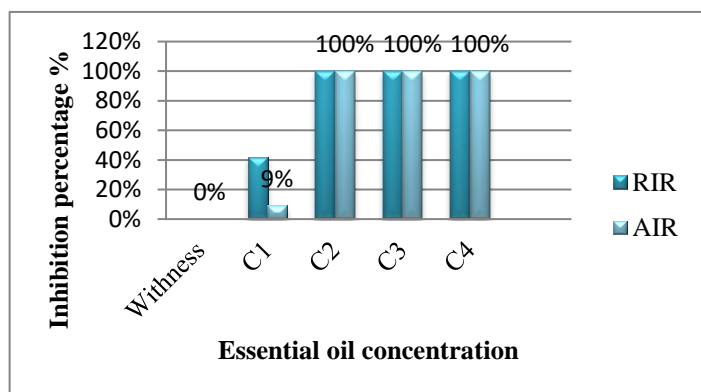


Figure 3. Inhibition of RIR and AIR recorded for Quinoa seeds treated with *F.vulgare* oil

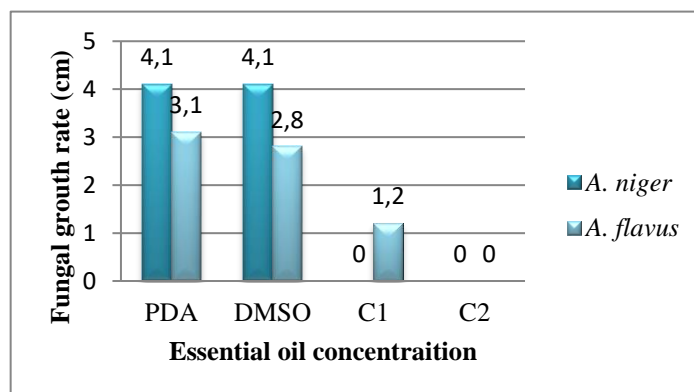


Figure 4. Mycelial diameter growth under the effect of *F.vulgare* oil

3. Results and Discussion

3.1. Chemical composition of essential oil

Analysis of the spectral data of the compounds detected 11 compounds with those of the standards cited in the databases, Nist and Willy (Table 1), whose alpha.-phellandrene (29.44), Dill ether (21.52) and D-Limonene (19.47), (-)-carvone (9.76) respectively are regarded major components.

Rahmani (1986) has reported that wild fennel seeds essential oil from the Azrou region (Middle Atlas, Morocco) were rich in hydrocarbon monoterpenes with a predominance of a-phellandrene (39%), limonene (21.4%) and a-pinene (17.6%). While, Shahat et al. (2012), has reported that the wild plants showed much higher level of limonene (84.49%). These variations may be related to the combined effect of many agents

comprising genetic factors and geographic origin as reported by several studies (Bahmani et al., 2015; Gholami Zali et al., 2018 ; Mustapha et al., 2020).

3.1. Allelopathic activity

The effect of essential oil concentrations on the final seed germination rate of the species tested which are shown in (figure1). We note that the percentage of inhibition of the aerial part is higher than the percentage of root inhibition for quinoa seeds at (C1) and it increases with increasing of concentration of the essential oil. The inhibitory effect is total for wheat seeds with different concentrations (figure 2, figure 3). These tests proved that our essential oil has an inhibitory effect on the 2 seeds germination tested; with relatively high inhibition rates.

In Previous studies basic research on the allelopathic potential of fennel (*Foeniculum vulgare*) seed at several

concentrations showed that this medicinal plant exhibited a significant inhibitory effect on the seed germination and seedling lengths of all examined weeds (Maryam et al., 2011). (Gilani et al. 2010) also reported that *F. vulgare* is one of the top 10 medicinal plants with the highest inhibitory effect against the germination of lettuce (*Lactuca sativa L.*) seeds .

3.2. Anti-fungal activity

The activity of different concentrations of essential oil of fennel on different fungal strains tested is revealed by the absence or presence of mycelial growth (figure 4).

According to (figure 4), it is observed that the inhibition rate has a fungicidal effect for the two fungal strains. It is increased with the increase of EO concentration.

Previous studies also found that fennel essential oils possessed an inhibitory effect against a wide range of fungi (Anwar et al., 2009, Singh et al., 2003) , which supported our results in the present study, indicating that essential oil of fennel was a potent fungal inhibitor.

4. Conclusions and Recommendations

This study has been devoted to determine the essential oil composition and to evaluate allelopathic and anti fungal activities.

analysis of the *F.vulgare* essential oil made it possible to identify 11 constituents, *F. vulgare* EO is dominated by alpha-phellandrene , Dill ether, D-Limonene and (-)-carvone.

F.vulgare essential oil exhibited a significant inhibitory effect on the seed germination of two examined weeds and on mycelial growth of the fungal strains.

This indicates that EO of *F. vulgare* could be regarded as a promising preservative in food industry in terms of having antifungal activity for food deterioration control, and may be promising alternative to synthetic herbicide.

It is recommended that other strains should be also used in order to test out their significance. However, in vivo studies must be performed to confirm this efficacy in vitro.

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