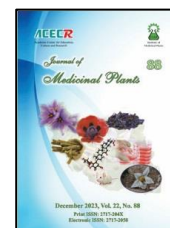




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### Research Article

## Antimicrobial and antioxidant activities of essential oils from seven species of *Amaryllidaceae* and *Brassicaceae*

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| ARTICLE INFO                                                                                                                                                                 | ABSTRACT                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                              |
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| <p><b>Keywords:</b></p> <p><i>Allium</i></p> <p>Antibacterial agents</p> <p>Antifungal agents</p> <p>Antioxidants</p> <p>Essential oils</p> <p><i>Fortuynia garcinii</i></p> | <p><b>Background:</b> Essential oils have been utilized for various purposes throughout history. These aromatic substances have become increasingly popular in alternative medicine, aromatherapy, and personal care products. <b>Objective:</b> In this study, essential oils from the aerial parts of four <i>Allium</i> species and three <i>Brassicaceae</i> members, namely <i>Fortuynia garcinii</i>, <i>Draba verna</i>, and <i>Thlaspi arvense</i> were evaluated for their antioxidant, antibacterial, and antifungal properties. <b>Methods:</b> The radical-scavenging properties were tested using DPPH assay. Antimicrobial activities were examined on nine standard pathogens: three Gram-positive bacteria including <i>Staphylococcus aureus</i>, <i>Bacillus cereus</i>, <i>Streptococcus pyogenes</i>, three Gram-negative bacteria including <i>Escherichia coli</i>, <i>Pseudomonas aeruginosa</i>, <i>Salmonella enterica subsp. enterica</i> and two fungi <i>Aspergillus fumigatus</i>, <i>Fusarium oxysporum</i> as well as the yeast <i>Candida albicans</i>. <b>Results:</b> The IC<sub>50</sub> values of antioxidant assay ranged from 124.66 to 155.04 µg/ml. <i>Allium zagricum</i> showed the best antioxidant effects with IC<sub>50</sub> of 124.66 µg/ml compared to standard vitamin E (IC<sub>50</sub> = 10.40 µg/ml). Similarly, the MIC values of 25-400 µg/ml with <i>Fortuynia garcinii</i> fruits-Zahedan were assessed as the best antimicrobial effects, while they were higher than the MIC values recorded for positive controls (0.06-16 for amikacin and 32-256 for clotrimazole). <b>Conclusion:</b> Essential oils extracted from <i>Allium zagricum</i> and <i>Fortuynia garcinii</i> can be prescribed for the treatment of oxidative stress-related and infectious diseases.</p> |

**Abbreviations:** FFH, *Fortuynia garcinii* Fruits-Zahedan; FFB, *Fortuynia garcinii* Fruits-Zabol; FLB, *Fortuynia garcinii* Leaves- Zabol; DVL, *Draba verna* L.; TAL, *Thlaspi arvense* L.; AIR, *Allium iranshahrii*; AAR, *Allium alamutense* Razyfard, Zarre; ACH, *Allium chrysantherum*; AZA, *Allium zagricum*; VIE, Vitamin E; AMK, Amikacin; CLT, Clotrimazole; DPPH, 2,2-Diphenyl-1-picrylhydrazyl; MIC, Minimum Inhibitory Concentration; MBC, Minimum Bactericidal Concentration; MFC, Minimum Fungicidal Concentration; PTCC, Persian Type Culture Collection; IBRC, Iranian Biological Resource Centre; UOZH, University of Zabol Herbarium; SD, Standard deviation; GC-MS, Gas Chromatography-Mass Spectrometry; EE, The essential oil extraction efficiency; DMSO, Dimethyl sulfoxide; ABTS, 2,2'-Azino-bis(3-ethylbenzothiazoline-6-sulfonic acid)

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## 1. Introduction

For many centuries, herbal products were the only available option for the prevention and treatment of many diseases in humans and animals. Plant extracts and essential oils contain valuable chemical compounds that have shown promising potential in the fields of medicine, food, cosmetics, and agriculture [1]. Essential oils are intricate combinations of fragrant substances that occur naturally in various parts of aromatic plants. They are typically colorless or light yellow liquids, soluble in lipid and organic solvents, and with molecular weights less than 300 Da [2, 3].

Developing synthetic preservatives and fragrances in various industries is more expensive compared to natural, plant-based essential oils while the latter could be valueless and even unsafe for human applications. Some chemicals found in manufactured fragrances include endocrine-disrupting activities like phenol and phthalates, and carcinogenic ingredients such as benzene derivatives [4, 5]. In addition, the increasing global tendency towards natural fragrances and additives, due to their safety and health benefits, has boosted the efforts to find natural options to replace synthetic fragrances and preservatives.

The antioxidant, antimicrobial, and antiparasitic activities of essential oils have been well documented and are thought to come from the presence of phenolic functional groups [6]. The essential oils exert their antimicrobial activity through different mechanisms with more efficacy on Gram-positive bacteria than Gram-negative ones [7]. The researchers put a lot of effort into discovering plant-based antioxidant and antimicrobial components all around the world. The antimicrobial and antioxidant activities of a huge number of plants have been investigated worldwide, but there is still a long

way to investigate many other valuable plant resources. Endemic plants that are found in limited phytogeographical, ecological, or political ranges are of critical importance. These invaluable plants are often unknown or inaccessible to many researchers. In addition, they are unique genetic pools of specific geographical regions and could be very important in different aspects. They may contain unique ingredients or characteristics but due to the small population size are prone to extinction, leading to the loss of all possible information and potentials.

The genus *Allium* L. (derived from the Greek word also meaning to avoid, due to the smell of garlic) was described by Carl Linnaeus in 1753 [8]. *Allium* (Amaryllidaceae) with around 1000 species is one of the largest genera in the monocotyledons distributed predominantly in the northern hemisphere [9]. Some of these species have been cultivated and used for centuries, not only as culinary ingredients but also for their medicinal properties. *Allium* is also the largest genus of petaloid monocots in Iran with around 153 species allocated to eight subgenera and 32 sections [10]. Members of this genus occupy many different habitats from sea shores, and the vicinity of salt lakes to montane and even subalpine areas [11]. A few *Allium* species such as *A. cepa*, *A. sativum*, and *A. tenuissimum* have been studied extensively for their chemical composition and antioxidant and antimicrobial properties [12-14]. However, there is no information regarding the biological properties of many endemic species of *Allium* from Iran. Four *Allium* species investigated in this study (*A. alamutense* Razyfard, *A. chrysanthrum* Zarre & R.M.Fritsch, *A. iranshahrii* R.M.Fritsch, and *A. zagricum* R.M.Fritsch) are narrow endemics with very limited distribution ranges in West and North West of Iran while distribution range of *A.*

*chrysanthemum* stretches from West of Iran to adjacent countries of Iraq, Turkey, Syria, and Jordan.

*Fortuynia* (Brassicaceae) contains only a single species, *F. garcinii* (Burm.f.) Shuttlew. which is endemic to Iran, Afghanistan, and western Pakistan (*Flora iranica*). the genus name and specific epithet refer to an Indonesian and a French botanist respectively. The plant is perennial, woody at base, glabrous, with simple, entire,  $\pm$  fleshy leaves and biarticulated broadly winged Siliculae. *Draba verna* L. (*Draba* is Greek for bitter, referring to the taste of the plant and *verna* is Latin for springtime when the plant blooms) is a small annual member of the family Brassicaceae with basal leaves, bifid petals and leafless stems growing widely around the world. *Thlaspi arvense* L. is also a small glabrous annual plant in the family Brassicaceae native to the Mediterranean region but becoming widespread almost all around the world. The generic name comes from a Greek word for cress and *arvense* is derived from the Latin word "arvum" meaning ploughed, pertaining to the cultivated and disturbed lands where it grows [15].

The objective of this study was to gain a background information on antioxidant and antimicrobial properties of four species of the genus *Allium* namely *A. Iranshahrii*, *A. Alamutense*, *A. zagricum* and *A. chrysanthemum* as well as three selected species from the Brassicaceae family including *Fortuynia garcinii*, *Draba verna* and *Thlaspi arvense*. Certainly, understanding the background knowledge and applicability of these unique

plants can provide valuable insights into their potential medicinal, nutritional or agricultural benefits.

## 2. Materials and Methods

### 2.1. Plant Material Collection

The wild plant species were collected from their natural habitats during flowering stage. The scientific names, geographical localities, herbarium codes, and characteristics of the examined plant species are provided in Table 1. The plant species were identified using botanical resources by the third and fourth authors, botanists at the Department of Biology, University of Zabol and the curator of the IBRC herbarium, respectively. A herbarium sample of each species was prepared and restored either at the UOZH in Zabol, Iran or IBRC herbarium located in Karaj, Iran (for details see Table 1). A photograph of each studied plant species, its habitat and detailed characteristics has been presented in Fig. 1. The targeted parts of the sampled plants were shade dried for a week, of which 100 g were converted to powder using an electronic machine.

### 2.2. Essential Oil Extraction

10 g of each powdered material was hydrodistilled for 4 h with 300 ml of distilled water, using a micro-clevenger type apparatus. The volatile fractions were extracted with 10 ml diethyl ether. The extract was dried with 1 g anhydrous magnesium sulfate. Finally, the resulting solution was condensed under a nitrogen stream.

**Table 1.** The scientific names, geographical distribution, localities, herbarium codes, and characteristics of the examined plant species

| No | Species                                                   | Family         | Herbarium code | Characteristics                                                                                                                                                       | Locality                                                                  | Distribution range                     | Used part        |
|----|-----------------------------------------------------------|----------------|----------------|-----------------------------------------------------------------------------------------------------------------------------------------------------------------------|---------------------------------------------------------------------------|----------------------------------------|------------------|
| 1  | <i>Allium iranshahrii</i><br>R.M.Fritsch                  | Amaryllidaceae | 1824           | Bulbous; Leaves 1-2, laminae broadly ovate to elliptic, upper side with deep furrows; tepals whitish to rose with a broad green to brown median vein, anthers carmine | W Azerbaijan: Piranshahr, Silveh, Mashkan, Chighidarreh mt. 2835m.        | NW Iran                                | Aerial parts     |
| 2  | <i>Allium alamutense</i><br>Razyfard, Zarre & R.M.Fritsch | Amaryllidaceae | 1616           | Bulbous; scape 2-5 cm long; leaves 2, laminae broadly canaliculate; Flowers ± broadly funnel-shaped                                                                   | Zanjan: Zanjan towards Gilvan, 5 km before Khanchaei Rahdarkhaneh . 2435m | NNW Iran                               | Aerial parts     |
| 3  | <i>Allium chrysanthrum</i><br>Boiss. & Reut.              | Amaryllidaceae | 1630           | Bulbous; scape 50-80 cm long; tepals yellow, narrowly linear-lanceolate, all flower parts of the innermost flowers often purple flushed to dark purple                | Ilam: Ilam towards Eyvan, Chovar, Reno forest park, 1464m                 | W Iran to N Iraq, S Turkey and Jordan. | Aerial parts     |
| 4  | <i>Allium zagricum</i><br>R.M.Fritsch                     | Amaryllidaceae | 1673           | Bulbous; leaf laminae up to 8 cm broad, leaves glossy yellowish-green; tepals lanceolate-triangular, pale lilac                                                       | Lorestan: N Khorramabad, Qalemorghi, Taf Mts. 2285m                       | W Iran (Zagros Mountains)              | Aerial parts     |
| 5  | <i>Fortuynia garcinii</i><br>(Burm.f.) Shuttlew.          | Brassicaceae   | UOZH 1505      | Perennial with, entire, ± fleshy leaves and, winged Siliculae                                                                                                         | Sistan and Baluchestan, Zabol, 25 km from Zabol towards Nehbandan 478m    | Iran, Afghanistan and Pakistan         | Leaves<br>Fruits |
| 6  | <i>Fortuynia garcinii</i><br>(Burm.f.) Shuttlew.          | Brassicaceae   | UOZH 1506      | Perennial with, entire, ± fleshy leaves and, winged Siliculae                                                                                                         | Sistan and Baluchestan, Zahedan 25 km from Zahedan towards Khash          | Iran, Afghanistan and Pakistan         | Fruits           |
| 7  | <i>Draba verna</i><br>L.                                  | Brassicaceae   | UOZH 1507      | Small therophyte with basal leaves, bifid petals and leafless stems                                                                                                   | Hamedan, Asadabad, Shamsabad village 1673m                                | Native to Eurasia                      | Whole plant      |
| 8  | <i>Thlaspi arvense</i><br>L.                              | Brassicaceae   | UOZH 1508      | Small glabrous therophyte with narrow and toothed stem leaves                                                                                                         | Hamedan, Asadabad, Shamsabad village 1673m                                | Cosmopolitan                           | Whole plant      |



**Fig. 1.** Photographs of the studied plant species and their habitat and some morphological details. A) *Allium chrysantherum*, with a close up of the inflorescence; B) *Allium alamutense*; C) *Allium iranshahrii*; D) *Allium zagricum*; E) *Draba verna*; F) *Thlaspi arvense*, with details of fruiting inflorescence; G) *Fortuynia garcinia*, showing also details of fruits and the sampling for this study

### 2.3. Antimicrobial Activity

Six Gram-positive and Gram-negative bacteria, two fungal strains and a yeast were tested for their susceptibility to the essential oils extracted from four species of *Allium* and three species of *Brassicaceae*. The standard strains of *Pseudomonas aeruginosa* (ATCC 10145), *Salmonella enterica subsp. enterica* (ATCC 19585), *Escherichia coli* (ATCC 25922), *Staphylococcus aureus* (NCTC 7445), *Streptococcus pyogenes* (ATCC 12204), *Bacillus cereus* (ATCC 14579), *Aspergillus fumigatus* (PTCC 5009), *Fusarium oxysporum* (CBS 620.87) and *Candida albicans* (ATCC 10231) were used in this *in vitro* antimicrobial assay. All microbial strains were brought from the PTCC, Karaj, Iran. The MIC, MBC and MFC values of the essential oils were quantified in sterile 96-well plates by employing microdilution method [16]. The stock solutions of each essential oil were prepared using DMSO as solvent at initial concentrations of 80000, 40000, 20000, 10000, 8000, 4000, 2000, 1000, 500, 250, 125, and 62.5 µg/ml. 20 µl of each stock solution was respectively added to the 1<sup>st</sup> to 12<sup>th</sup> wells in each row of the 96-well plate. Then, 100 µl of microbial suspensions and 80 µl of liquid medium were added to all wells. The final concentrations of 8000, 4000, 2000, 1000, 800, 400, 200, 100, 50, 25, 12.5, 6.25 µg/ml of each sample were obtained in each row. The 96-well plates were incubated in 37 °C for 24 h. Amikacin and clotrimazole were applied as positive controls in this study. DMSO was also used as a negative control. While its final concentration was 10% (v/v) in each well, no inhibitory activity was observed with it. Finally, the wells with the least concentration that showed no visual turbidity were identified as the MIC. The MBC and MFC were determined by reculturing the contents of the transparent wells

in MIC phase with the lowest concentration where all microorganisms were disappeared.

### 2.4. Antioxidant Capacity and IC<sub>50</sub> Determination

The antioxidant capacity of essential oils was estimated according to their capacity to scavenge DPPH stable free radicals, as outlined in our previous publications [17, 18]. Initially, methanolic solutions of 50, 100, 200, and 400 µg/ml were created using each essential oil. Then, 3 ml of a fresh DPPH solution (0.004 % w/v) was added to 1 ml of each methanolic solution. The mixture was allowed to rest for 30 minutes in dark at ambient temperature to complete the reaction and discolor DPPH. Finally, a UV spectrophotometer (UV-2100 RAY Leigh) was used to determine the absorbance of each solution at 517 nm. The absorbance of the DPPH solution was read as blank. The equation  $I\% = [(A_0 - A_s)/(A_0)] \times 100$  was employed to detect the DPPH scavenging effect in which I % is inhibition percentage, A<sub>0</sub> is the absorbance of the blank and A<sub>s</sub> is the absorbance of the test sample. Finally, the antioxidant activity was reported as IC<sub>50</sub> values (µg/ml), indicating the required concentration of essential oil to exert 50 % inhibition. A similar procedure was followed for vitamin E which was used as a positive control in this study.

### 2.5. Statistical Analyses

The tests were performed three times to ensure the validity of the results, with the reported values representing the mean of these three independent trials ± SD. No SD was observed in susceptibility antimicrobial tests. Graphs were constructed using EXCEL software, and the data were statistically analyzed using version 16 of the SPSS software.

3. Results

Volatile oils of all seven species were extracted using steam distillation. The essential oil extraction efficiency was calculated according to the following formula:

$$EE\ (\%) = [m_o\ (g) / m_p\ (g)] \times 100$$

Where  $m_o$  and  $m_p$  are the mass of the obtained essential oil and the plant organ. The oil extraction yields were reported in Table 2.

All plants were evaluated for their possible antioxidant and antimicrobial properties. For this purpose, hydrogen-donating ability of essential

oils were investigated against DPPH free radicals, as shown in the Fig. 2. Moderate antioxidant effects were observed with them. *Allium* species were more successful in inhibiting radicals than *Fortuynia*, *Draba* and *Thlaspi* species. The  $IC_{50}$  values of *Allium* species were in the limited range of 124.66-128.73  $\mu\text{g/ml}$ . In *F. garcinii* samples, Zabol fruit had better effects than its Zahedanian counterpart. DVL and TAL had intermediate antioxidant effects. The  $IC_{50}$  values of essential oils were higher than those of vitamin E as standard (10.40  $\mu\text{g/ml}$ ).

Table 2. The essential oil extraction efficiency of samples

| Oils   | FFH   | FFB   | FLB   | DVL   | TAL   | AIR   | AAR   | ACH   | AZA   |
|--------|-------|-------|-------|-------|-------|-------|-------|-------|-------|
| EE (%) | 0.040 | 0.745 | 0.208 | 0.559 | 0.526 | 0.161 | 0.040 | 0.140 | 0.035 |

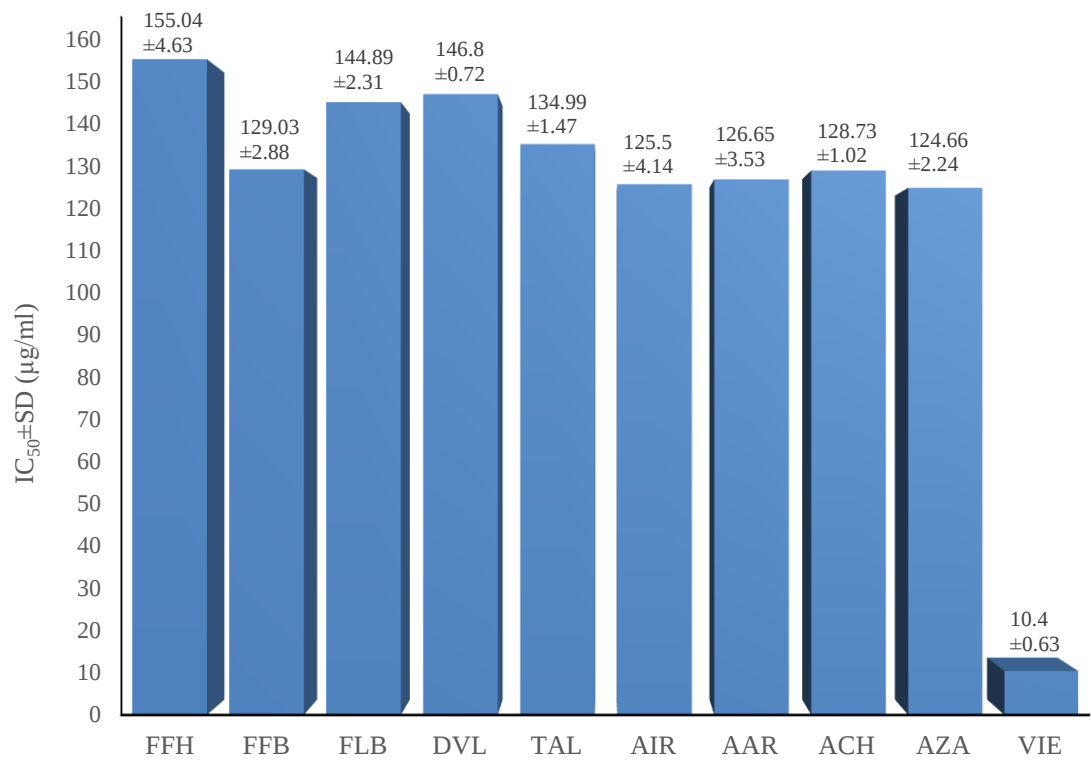


Fig. 2. DPPH free radical scavenging activity of essential oils

Antimicrobial potential of all essential oils was assessed against nine pathogenic bacterial and fungal strains from different genera. The MIC, MBC and MFC values of effective samples were recorded in the range of 25-8000 µg/ml. As shown in Table 3, no inhibitory activity was observed at the highest selective concentration (8 mg/ml) against *B. cereus* and *C. albicans*. Essential oil of FLB was the only effective specie on *S. pyogenes*. All three samples of *Fortuynia*

could inhibit the growth of *S. aureus* strain. AZA and DVL affected only *P. aeruginosa* and *A. fumigatus*, respectively. No antibacterial activity was observed with TAL against tested pathogens. AMK and CLT as positive controls present better inhibitory effects than essential oils with MICs of 0.06-16 and 32-256 µg/ml, respectively. Major essential oils (6 out of 9) could inhibit the growth of *S. aureus* and *A. fumigatus* strains with MICs of 400-8000 and 200-8000 µg/ml, respectively.

**Table 3.** Antimicrobial properties of essential oils and standards

|                      |                  | Essential oils |      |      |      |     |      |      |      | Standards |      |     |
|----------------------|------------------|----------------|------|------|------|-----|------|------|------|-----------|------|-----|
| Strains              |                  | FFH            | FFB  | FLB  | DVL  | TAL | AIR  | AAR  | ACH  | AZA       | AMK  | CLT |
| <i>S. aureus</i>     | MIC <sup>a</sup> | 400            | 2000 | 2000 | ND   | ND  | 2000 | 8000 | 2000 | ND        | 0.5  | -   |
|                      | MBC <sup>a</sup> | 800            | 2000 | 2000 | ND   | ND  | 2000 | 8000 | 2000 | ND        | 1    | -   |
| <i>B. cereus</i>     | MIC              | ND             | ND   | ND   | ND   | ND  | ND   | ND   | ND   | ND        | 0.5  | -   |
|                      | MBC              | ND             | ND   | ND   | ND   | ND  | ND   | ND   | ND   | ND        | 0.5  | -   |
| <i>S. pyogenes</i>   | MIC              | ND             | ND   | 4000 | ND   | ND  | ND   | ND   | ND   | ND        | 1    | -   |
|                      | MBC              | ND             | ND   | 4000 | ND   | ND  | ND   | ND   | ND   | ND        | 4    | -   |
| <i>E. coli</i>       | MIC              | 100            | 4000 | ND   | ND   | ND  | 4000 | ND   | ND   | ND        | 16   | -   |
|                      | MBC              | 100            | 4000 | ND   | ND   | ND  | 4000 | ND   | ND   | ND        | 16   | -   |
| <i>P. aeruginosa</i> | MIC              | 25             | ND   | 4000 | ND   | ND  | 4000 | ND   | 4000 | 8000      | 0.06 | -   |
|                      | MBC              | 25             | ND   | 4000 | ND   | ND  | 4000 | ND   | 4000 | 8000      | 0.06 | -   |
| <i>S. enterica</i>   | MIC              | ND             | ND   | ND   | ND   | ND  | 2000 | ND   | 2000 | ND        | 0.5  |     |
|                      | MBC              | ND             | ND   | ND   | ND   | ND  | 2000 | ND   | 4000 | ND        | 1    |     |
| <i>A. fumigatus</i>  | MIC              | 200            | ND   | 2000 | 2000 | ND  | 1000 | 8000 | 2000 | ND        | -    | 32  |
|                      | MFC              | 200            | ND   | 2000 | 2000 | ND  | 2000 | 8000 | 2000 | ND        | -    | 32  |
| <i>F. oxysporum</i>  | MIC              | 200            | ND   | 4000 | ND   | ND  | 2000 | ND   | 4000 | ND        | -    | 256 |
|                      | MFC <sup>a</sup> | 200            | ND   | 4000 | ND   | ND  | 2000 | ND   | 4000 | ND        | -    | 512 |
| <i>C. albicans</i>   | MIC              | ND             | ND   | ND   | ND   | ND  | ND   | ND   | ND   | ND        | -    | 256 |
|                      | MFC              | ND             | ND   | ND   | ND   | ND  | ND   | ND   | ND   | ND        | -    | 512 |

#### 4. Discussion

The primary treatment of diseases was mainly based on the use of medicinal plants, which is still thriving despite the development of synthetic drugs. For this purpose, the plant itself is

consumed directly or prescribed in the forms of extract and essential oil. These extracts and essential oils can be obtained from aerial and underground organs of plant such as flowers, fruits, leaves, peels, seeds, roots and bark. In this

study, antioxidant and antimicrobial potentials of essential oils of three *Brassicaceae* members and four *Allium* species were assessed against DPPH and a variety of microbial pathogens. No biological investigation was observed on *F. garcinii* despite its use in the treatment of spasm sedative, menstruation additive, and migraine in traditional medicine [19]. The water and ethyl acetate extracts of *D. tortuosa*, a plant closely related to *F. garcinii* and native to Pakistan, exhibited significantly greater antioxidant activity than that of *F. garcinii*. Moreover, its extracts were found to possess potent antibacterial properties effective against a wide range of bacteria and fungi [20]. Antimicrobial and antioxidant data from *D. verna*, *T. arvense*, *A. iranshahrii*, *A. chrysanthemum*, *A. zagricum* and *A. alamutense* was also not found for comparison. While none of the investigated *Allium* species in this study have been subjected to antioxidant and antimicrobial examinations, several other species have been extensively studied for their biological activities. *A. rubellum* which is phylogenetically related to *A. iranshahi* [21] showed significant free radical scavenging activity in ABTS and DPPH assays and good antibacterial activity against *E. coli* and *S. aureus* using disc diffusion method [22]. This is in congruence with the results of this paper. The *Allium* species that are phylogenetically close to *A. zagricum*, *A. alamutensis*, and *A. chrysanthemum* have not been studied yet, however, the antimicrobial effect of the widely used *Allium* species such as *A. sativum*, *A. cepa*, and *A. stipitatum* has been demonstrated against various bacteria and fungi, showing wide spectra of bactericide and fungicide properties [23, 24]. The similar antioxidant effects were observed with tested *Allium* species, which was probably to extract similar components during the steam distillation process. Various antimicrobial effects

were observed with the tested species, it can be due to the difference in the species genetics and/or climatic conditions [25]. Plant organ, extraction method, extraction solvent, sensitivity of bacteria, concentration of the extract, and the growth medium are other variables influencing antimicrobial properties [26]. Satyal *et al.* evaluated the chemical compositions of *A. vineale* and *A. sativum* volatile oils [27]. Their GC-MS analysis proved the presence of a wide spectrum of sulfur-containing compounds (>90 %) such as disulfide, 3-methyl/2,4-dimethyl/3,4-dimethylthiophenes, 1,2-dithiolane/dithiolene, [1-propenyl] 2-thiopent-3-yl/propyl 4-thiohept-2-en-5-yl/dimethyl/diallyl sulfides/disulfides, allyl/allyl methyl/methyl propyl/methyl methylthiomethyl/allyl methylthiomethyl/1-propenyl/1-propenyl propyl sulfides/disulfides, diallyl/methyl propyl/methyl 1-propenyl/dimethyl/allyl methyl/allyl propyl/allyl 1-propenyl trisulfides, 4-methyl-1,2,3-trithiolane, 2/3-vinyl-4*H*-1,2/1,3-dithiines, 1,2,3-trithia-4-cyclohexene, allicin, methyl (methylsulfinyl)methyl sulfide, 5-methyl-1,2,3,4-tetrathiane, 1,4-dihydro-2,3-benzoxathiin 3-oxide, diallyl/dimethyl/allyl methyl tetrasulfide, 4-methyl-1,2,3,5,6-pentathiepane and cyclooctasulfur. It appears that the antimicrobial properties of *Brassica* and *Allium* families originate from their volatile sulfur compounds [28, 29]. Blocking effects of diallyl trisulfide, diallyl disulfide, dimethyl trisulfide, diallyl tetrasulfide and allyl isothiocyanate, which are among the compounds present in *Allium* and *Brassica* species, have been proven against *S. aureus*, *E. coli*, *E. aerogenes*, *L. mesenteroides*, *P. pentosaceus*, *L. plantarum*, *C. albicans*, *C. utilis*, *S. cerevisiae*, *P. membranifaciens*, *Z. bisporus* and *Z. rouxii* with MICs of from 1 to above 1000 ppm [30]. These components react with thiol (SH) groups of cell

wall proteins to inhibit the growth of microorganisms [31]. Antibacterial and antifungal potentials of ethanolic extracts of *Allium sativum* L. from China and Turkey were investigated on bacterial species including *Enterobacter*, *Bacillus*, *Listeria*, *Enterococcus*, *Salmonella*, *Klebsiella*, *Escherichia*, *Pseudomonas* and *Staphylococcus* as well as *C. albicans* yeast [32]. Inhibition zones were ranged from 7 to 30 mm; Turkish samples was more effective than Chinese samples in killing microorganisms. Antimicrobial effects of essential oils of an *A. sativum* and three *A. cepa* (red, yellow and green) in the concentration range of 50-500 ml/l were assessed on *S. Enteritidis*, *S. aureus*, *P. cyclopium*, *F. oxysporum* and *A. niger* strains [33]. *A. sativum* and green *A. cepa* were identified as the best and worst antimicrobial agents, respectively. The lowest sensitivities were observed with essential oils against *F. oxysporum* and *S. aureus*.

A direct relationship is usually observed between antimicrobial properties and antioxidant potentials of compounds. The antioxidant ability of plants is mainly dependent on their phenolic content. These phenolic compounds prevent oxidative stress via reduction and neutralization of free radicals, coordination to metal ions, and scavenging singlet and triplet oxygen [34]. Phenolic compounds play an important role in the antimicrobial properties of plants. They kill microorganisms by destroying cell walls and proteins, membrane and enzyme dysfunction, and disrupting DNA and RNA replication [35]. It is found that the antibacterial properties of essential oils can be caused by the following factors: 1) The hydrophobic property of the essential oil allows it to penetrate the bacterial cell membrane and causes the release of ions and other internal components of the cells; 2) Phenolic derivatives present in the essential oils such as carvacrol, eugenol, and thymol destroy the cytoplasmic membrane of the cell and interfere with the proton and electric current; 3) Some components

especially cinnamaldehyde can attach to cell proteins such as decarboxylase to prevent its vital function; 4) The presence of alphasitriptin and molecules containing SH group is effective in inhibiting the growth of many microorganisms [36].

## 5. Conclusion

The antioxidant capacity and antimicrobial activity of essential oils of four *Allium* species and *F. garcinia*, *D. verna*, and *Th. arvense* (Brassicaceae) from Iran were determined for the first time. Moderate antioxidant and moderate to excellent antimicrobial effects were observed with them. Although AZA showed the best antioxidant effects, no antimicrobial effect was observed with it on tested microbial strains. FFH showed more effective and broad-spectrum antimicrobial effects compared to others. Essential oil of FFH can be introduced as a potent antimicrobial agent, its inhibitory effects may be ameliorated by climate change.

## Author contributions

H. B. supervised the data collection and prepared the final version of the manuscript. M. D. and A. D. contributed to the plant sampling and identification as well as manuscript preparation. N. M. prepared the essential oils. Z. E. collected the laboratory data. All authors have approved the final version of the manuscript.

## Conflicts of interest

The authors declare no conflict of interest associated with this manuscript. The authors alone are responsible for the content of the paper.

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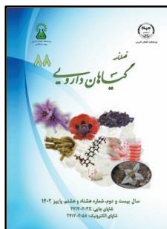
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## مقاله تحقیقاتی

## فعالیت‌های ضد میکروبی و آنتی اکسیدانی اسانس‌هایی از هفت گونه نرگسیان و شب بویان

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## چکیده

## اطلاعات مقاله

کل واژگان:

*Allium*

عوامل ضدباکتریایی

عوامل ضدقارچی

آنتی اکسیدان‌ها

اسانس‌ها

*Fortuynia garcinia*

**مقدمه:** اسانس‌ها در طول تاریخ جهت مصارف مختلفی مورد استفاده قرار گرفته‌اند. این مواد معطر در طب جایگزین، رایحه درمانی و محصولات مراقبت شخصی به طور فزاینده‌ای محبوب شده‌اند. **هدف:** در این مطالعه، اسانس‌هایی از اندام‌های هوایی چهار گونه *Allium* و سه گونه *Brassicaceae* شامل *Fortuynia garcinii*، *Draba verna* و *Thlaspi arvense* از نظر خواص آنتی اکسیدانی، ضدباکتریایی و ضدقارچی مورد ارزیابی قرار گرفتند. **روش بررسی:** خواص مهارکنندگی رادیکالی با استفاده از آزمون DPPH مورد آزمایش قرار گرفت. فعالیت ضد میکروبی اسانس‌ها علیه نه عامل بیماری‌زای استاندارد: سه باکتری گرم-مثبت شامل *استافیلوکوکوس اورئوس*، *باسیلوس سرئوس*، *استرپتوکوکوس پیوژنز*، سه باکتری گرم-منفی شامل *شریشیا کلی*، *سودوموناس آئروژینوزا*، *سالمونلا انتریکا* زیرگونه *انتریکا* و دو قارچ *آسپرژیلوس فومیگاتوس*، *فوساریوم اکسی پروم* و همچنین مخمر *کاندیدا آلبیکنس* مورد بررسی قرار گرفت. **نتایج:** مقادیر  $IC_{50}$  سنجش آنتی اکسیدانی در گستره ۱۲۴/۶۶ تا ۱۵۵/۰۴ میکروگرم بر میلی لیتر بودند. *Allium zagricum* با  $IC_{50}$  ۱۲۴/۶۶ میکروگرم بر میلی لیتر بهترین اثرات آنتی اکسیدانی را در برابر ویتامین E استاندارد (با  $IC_{50}$  ۱۰/۴۰ میکروگرم بر میلی لیتر) نشان داد. به همین ترتیب، مقادیر MIC ۲۰۰-۴۰۰ میکروگرم بر میلی لیتر به عنوان بهترین اثرات ضد میکروبی از میوه‌های *Fortuynia garcinii*-زاهدان کسب گردید، درحالیکه از مقادیر MIC ثبت شده برای کنترل‌های مثبت (۱۶-۰/۰۶) برای آمیکاسین و ۲۵۶-۳۲ برای کلوتریمازول بالاتر بودند. به ترتیب بهترین اثر آنتی اکسیدانی و ضد میکروبی را نشان دادند. **نتیجه‌گیری:** اسانس‌های مستخرج از *Allium zagricum* و *Fortuynia garcinii* را می‌توان در درمان بیماری‌های عفونی و مرتبط با استرس اکسایشی تجویز کرد.

**مخفف‌ها:** FFH: میوه‌های *Fortuynia garcinii*-زاهدان؛ FFB: میوه‌های *Fortuynia garcinii*-زابل؛ FLB: برگ‌های *Fortuynia garcinii*-زابل؛ DVL: *Draba verna* L.؛ TAL: *Thlaspi arvense* L.؛ AIR: *Allium iranshahrii*؛ AAR: Zarre، Razafard، *Allium alamutense*؛ ACH: *Allium chrysantherum*؛ AZA: *Allium zagricum*؛ VIE: ویتامین E؛ AMK: آمیکاسین؛ CLT: کلوتریمازول؛ DPPH: ۲،۲-دی فنیل-۱-پیکریل هیدرازیل؛ MIC: حداقل غلظت بازدارندگی؛ MBC: حداقل غلظت کشندگی باکتریایی؛ MFC: حداقل غلظت کشندگی قارچی؛ PTCC: مرکز منطقه‌ای میکروارگانیسم‌های ایران؛ IBRC: مرکز منابع زیستی ایران؛ UOZH: هرباریوم دانشگاه زابل؛ SD: انحراف استاندارد؛ GC-MS: کروماتوگرافی گازی-طیف سنجی جرمی؛ EE: راندمان استخراج اسانس؛ DMSO: دی‌متیل سولفوکسید؛ ABTS: ۲،۲-آزینو-بیس(اتیل بنزوتیازولین-۶-سولفونیک اسید)

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