



Antifungal and allelopathic activities of aqueous and methanolic extracts from *Ephedra alata* aerial parts

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Abstract

Although bioactivities of *Ephedra alata* have been widely described, most of them are related to its anticancer or antioxidant potencies while data related to its antifungal and allelopathic properties are still very scarce. Thus, the aim of this study is to determine the antifungal activity of the aqueous and methanolic extracts from *Ephedra alata* aerial parts and to assess their allelopathic potential. The antifungal potential was investigated qualitatively and quantitatively using disc diffusion and microdilution methods, while germination seed test was used to assess allelopathic activity. Antifungal results showed that all fungus strains were inhibited by aqueous and methanolic extracts in a dose-dependent manner. The aqueous extract showed the highest inhibition power on conidial germination of *A. fumigatus*. The methanolic extract exhibited strong antifungal activity against *A. niger* and *A. flavus*. Regarding the allelopathic effect, dose-dependent phytotoxicity activity against *Triticum aestivum*, *Linum usitatissimum*, and *Trigonella foenum-graecum* was observed with both of our extracts. Precisely, data demonstrated that the methanolic extract is more phytotoxic than the aqueous one. The obtained results are of interest for the potential use of *E. alata* extracts as a natural antifungal and as a biological herbicide.

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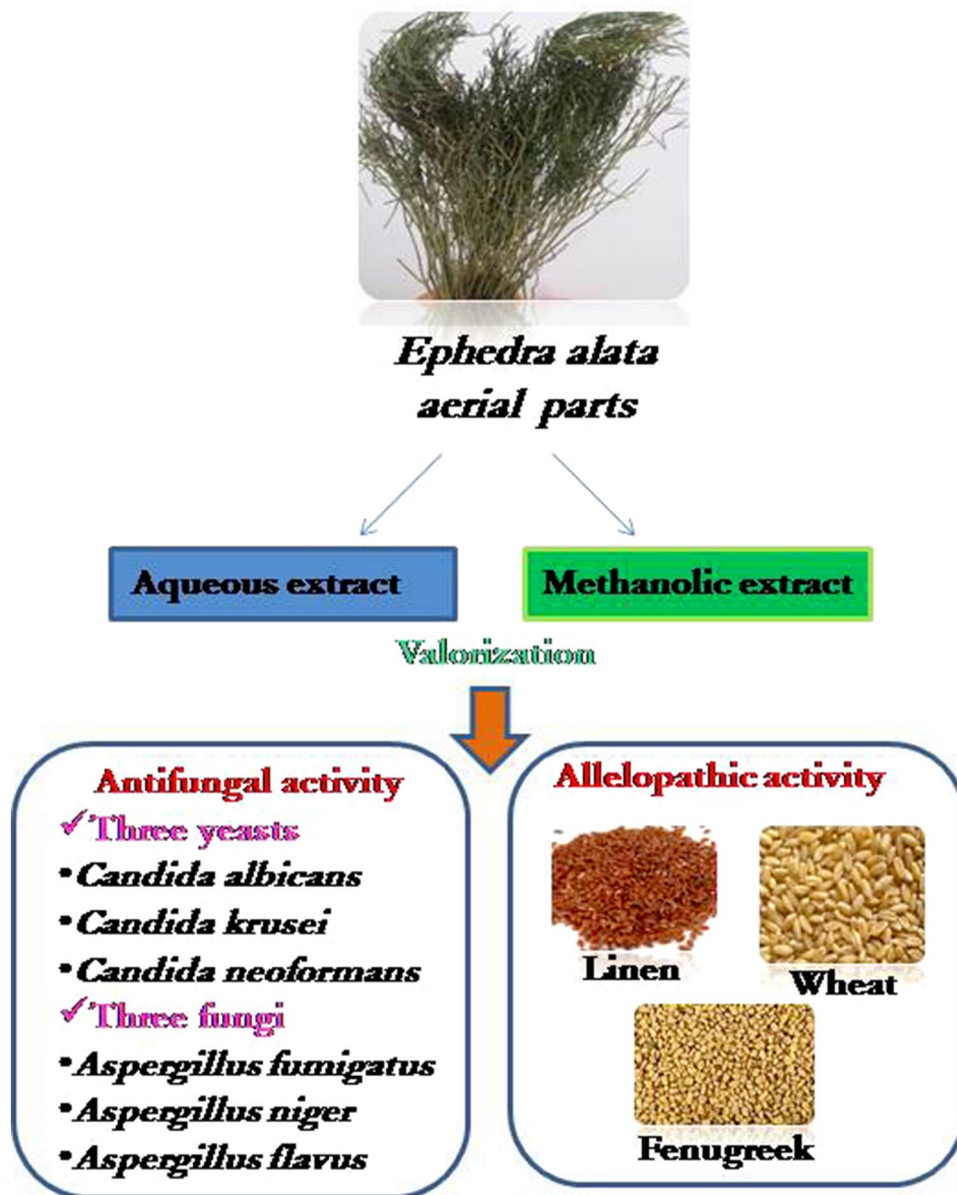
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Graphical abstract



Keywords *Ephedra alata* · Aqueous extract · Methanolic extract · Biological control · Antifungal *A. fumigatus* · *A. niger* · *A. flavus* · Allelopathic

Introduction

In recent years, there has been an increased interest in obtaining natural antifungals, mainly due to the etiology of the contamination of several types of food, including fruits, vegetables, cereals, meats, feed products (Gerez et al. 2013; Saladino et al. 2016), dried spices, aromatic herbs powder (Akpo-Djènontin et al. 2018), nuts (da Cunha et al. 2018), sweet pepper (Gambacorta et al. 2019), and dried date palm

fruit, by molds, which contribute to the destruction of nearly 30% of crop yields (Saladino et al. 2016). The deterioration of food and agricultural products has been attributed to mycotoxin contamination produced by various toxigenic fungi belonging to the genera *Aspergillus*, *Fusarium*, and *Penicillium* (Taheur et al. 2019). Aflatoxin B1 and ochratoxin A are considered among the most harmful mycotoxins (Taheur et al. 2019). Aflatoxin B1, which are mainly produced by *Aspergillus flavus*, *A. parasiticus*, and *A. nomius*,

are reported to be hepatotoxic, immunosuppressive, teratogenic, mutagenic, and carcinogenic (Sangmanee and Hongpattarakere 2014; Kabak 2016). Ochratoxin A is mainly produced by *A. ochraceus*, *A. carbonarius*, *A. niger*, and *Penicillium verrucosum* (De Bellis et al. 2015). It is well documented to induce many toxic properties such as nephrotoxicity, hepatotoxicity, immunotoxicity, embryotoxicity, teratogenicity, and carcinogenicity (Radić et al. 1997; Jo et al. 2016). Many chemical (synthetic antifungal products) and physical (filtration, adsorption, electric pulses, irradiation, etc.) strategies were applied to increase crop safety by preventing fungal contamination and reducing mycotoxin levels (Aziz et al. 2007; Adebayo and Aderiyé 2011). Nevertheless, these methods have some limitations including the accumulation of chemical residues in food and environment as well as the phenomena of mutation of fungi (Adebayo and Aderiyé 2011; Zhu et al. 2015). Currently, there has been increased interest in the use of plants and their derivatives as biological fungal control due to their safety and prevalence in nature (Sangsilá et al. 2016).

The yield and quality of cultivated plants may be negatively affected by climatic factors, pests, weeds, and diseases (Özen et al. 2017). Weed control is among the major obstacles in improving the world's agricultural efficiency and is considered as an important factor that affects crop quality and causes enormous economic loss in agricultural production (Patterson 1995). Recently, the overuse of chemical and synthetic herbicides has provoked herbicide-resistant weed phenomena and affected human health and the environment. Therefore, intensive research has been conducted to search for an alternative based on plant compounds that can reduce weed growth via allelopathy (Isman 2000).

Thus, many plants exhibit allelopathic power because they release a great variety of chemical substances generically named allelochemicals, which can affect positively or negatively the life of other coexisting plants and animals (Bouwmeester et al. 2003). Allelochemicals include a variety of secondary metabolites, especially phenolic compounds, flavonoids, alkaloids, and terpenoids (Reigosa et al. 2013; Areco et al. 2014). The allelopathic compounds can lead to strong inhibition of weed seed germination, insect, bacterial, and fungal influences (Duke et al. 2000). Therefore, the use of plants with allelopathic metabolites to develop biopesticides can be an effective tool for sustainable agriculture by reducing the use of synthetic pesticides.

Ephedra is a genus of the Ephedraceae family, with approximately 67 species distributed in arid and desert environments in the North Hemisphere, Asia, America, the southern part of Europe, and North Africa (Xie et al. 2013). *Ephedra alata*, a species of *Ephedra*, is commonly known in Tunisia as “Alenda” and widely distributed on the steppes and the deserts of the southern regions (Nabli 1991; Chaieb and Boukhris 1998). The decoction of

E. alata aerial parts is often used in traditional medicine to alleviate nasal stuffiness, digestive disorders, and respiratory diseases as well as a treatment for bacterial and fungal infections after abortion (Nawwar et al. 1984; Gupta et al. 2008). Recently, studies performed with different *E. alata* extracts have demonstrated numerous biological activities, including anticancer (Sioud et al. 2020a), anti-inflammatory (Soumaya et al. 2020), antidiabetogenic (Lamine et al. 2019), anti-obesity (Kim et al. 2014), antiviral (Soltan and Zaki 2009), antifungal (Al-Qarawi et al. 2013), antioxidant, antibiofilm, and antibacterial (Dbeibia et al. 2022) properties.

Furthermore, several phytochemical compounds such as alkaloids, phenolic acids, flavonoids, proanthocyanidins, tannins, saponins, reducing sugars, cardiac glycosides, and fatty acids have been reported to be present in the aerial parts of *E. alata* (Ibragic and Sofić 2015; Sioud et al. 2020a, 2020b; Dbeibia et al. 2022).

The goals of this study were to evaluate the antifungal activity of aqueous and methanolic extracts of *E. alata* upon three yeasts and three toxigenic fungal strains and to study their allelopathic effect on *Triticum aestivum*, *Linum usitatissimum*, and *Trigonella foenum-graecum*. To the best of our knowledge, this is the first report on the antifungal and allelopathic potencies of *E. alata* aerial parts.

Material and methods

Plant material

Ephedra alata was collected during the flowering season, from Kef Derbi area (latitude: 34°41', longitude: 9°29'), Governorate of Gafsa (Tunisia) in March 2021. The whole aerial parts of the plant were rinsed with deionized water, air-dried in shade, and finely ground.

Extracts preparation

To prepare the aqueous extract, 10 g of powder samples was mixed with 100 mL of distilled water and stored at room temperature overnight. Samples were then filtered under reduced pressure and finally lyophilized. For the methanolic extract, the same amount of powder was immersed in the methanol (Merck; 97%), for 48 h at room temperature. Then, the mixture was evaporated to dryness under reduced pressure in a rotary evaporator (Hei-vap Presición HL G3 from Heidolph Technologies) at 45 °C, to remove the solvent. Both crude extracts were stored in amber bottles at 4 °C until further use.

Antifungal activity

The antifungal activity was tested against three yeasts (*Candida albicans* ATCC 90028, *Candida krusei* ATCC 6258, and *Candida neoformans* ATCC 14116) and three fungal strains (*Aspergillus fumigatus* ATCC 204305, *Aspergillus niger* 15UA006, and *Aspergillus flavus* 15UA005).

The anticandidal activity of the extracts was evaluated as previously described by Rios and Recio (2005). All the yeasts were grown in Sabouraud Chloramphenicol broth (Oxoid, Basingstoke, Hampshire, UK) at 25 °C for 24 h, and suspensions were adjusted to 0.5 McFarland standard turbidity. Afterwards, 100 µL of each precultured suspension was spread onto plates containing Sabouraud Chloramphenicol agar (SCA) (Oxoid, Basingstoke, Hampshire, UK). Sterile filter paper discs (6 mm in diameter) were impregnated with 20 µL of the different extracts and placed on agar. The treated plates were stored for 1 h at 4 °C and then incubated for 24 h at 25 °C.

In addition, the anticandidal activity of *E. alata* extracts was evaluated quantitatively by broth microdilution method as described by CLSI (2018). Briefly, each extract was serially diluted in Sabouraud broth (Oxoid, Basingstoke, Hampshire, UK) to obtain a final concentration ranging from 0.05 to 25 mg/mL in 96-well U-bottomed polystyrene plates. Then, 10 µL of each candidal suspension (0.5 McFarland) was added to each well and the microplates were incubated at 25 °C for 24 h. Minimal inhibitory concentration (MIC) value was defined as the lowest concentration of extracts that had no visible *Candida* growth. The minimal fungicidal concentration (MFC) was determined by spotting 10 µL medium of each well with no visible *Candida* growth onto a Sabouraud agar (Oxoid, Basingstoke, Hampshire, UK) plate and incubating at 25 °C for 24 h. MFC was defined as the lowest concentration of compounds at which all *Candida* was destroyed.

In the other hand, fungal strains were grown in SCA for 7 days at 25 °C. Spore suspension was prepared in sterile peptone water (0.1% w/v) supplemented with one to two drops of Tween 80 and adjusted to 10⁶ spores/mL. Antifungal activity of aqueous and methanolic extracts was evaluated as described by Cortés-Zavaleta et al. (2014) with slight modifications. Aqueous or methanolic extracts (10%, v/v) were mixed with SCA at 45 °C, poured into Petri dishes (20 mL per plate) and kept standing overnight at room temperature for media solidification. Then, 10 µL of mold spore suspension (10⁶ spores/mL) of the different fungi was spotted onto the center of the surface of the agar layer in each plate. Control SCA plates were mixed with sterile distilled water or dimethyl sulfoxide (10%, v/v) in the same proportions and inoculated. Experiments were repeated in triplicate. All the plates were incubated aerobically at 25 °C. Diameters of the mold growth were measured daily for

5 days, and the percentage of mold growth inhibition was calculated according to the following formula (1):

$$PI = [(Dc - DT) / Dc] * 100 \quad (1)$$

where PI is the percentage of growth inhibition and DT and DC are the diameter of mycelial growth in treated and control plates, respectively.

Allelopathic activity

To test the direct or indirect biochemical interactions, positive or negative, of the aqueous and methanolic extracts of *E. alata* aerial parts, three types of target seeds were used: *Triticum aestivum*, *Linum usitatissimum*, and *Trigonella foenum-graecum*. More specifically, the negative or positive effects of each indicated extract of *E. alata* on the kinetics of germination of these seeds were tested according to the protocol adopted by Zairi et al. (2020). The germination was carried out under the temperature and illumination conditions of the laboratory. The three types of seeds studied were germinated in Petri dishes on two layers of filter paper moistened with 5 mL of distilled water as a control treatment or with 5 mL of aqueous or methanolic extracts in different concentrations (1%, 0.5%, and 0.25%). Each box contained 20 seeds, and for each treatment three repetitions were performed. Germination monitoring lasted 1 week, with the seeds that sprouted being enumerated every day. The percentage of germination is calculated according to the following formula (2):

$$\% G = (NGG / NTG) * 100 \quad (2)$$

%G: percent of germination (%).

%IG: percent of germination inhibition = 100 – %G.

NGG: number of seeds germinated in the presence of water or extract.

NTG: total number of seeds.

Statistical analysis

MIC and MFC values obtained from the anticandidal activity measurement were the most frequently observed for each *Candida* strain.

Experiments were performed in triplicate, and the results are presented as data average ± standard deviation (SD).

Statistical analyses were performed with one-way analysis of variance (ANOVA) with one-factor comparisons by Tukey's test for the allelopathic activity and by Duncan's test for the antifungal activity by using GraphPad Prism software (Prism version 5.04). A *p* value less than 0.05 was considered statistically significant, and less than 0.001 was considered highly statistically significant.

Results

Our present work shows clearly that using the aqueous and methanolic extracts obtained from *E. alata* aerial parts achieved good results in inhibiting the yeast and conidial germination associated with the deterioration of food and agricultural products. Also, both extracts can inhibit seedling growth in a dose-dependent manner, making them useful for eco-friendly, biocontrol program management.

Antifungal activity

Antifungal activity of extracts against *Candida* strains

Results of the antifungal activity of *E. alata* extracts are expressed as diameters of the inhibition zones as well as MICs and MFCs (Table 1). The aqueous extract was active only toward *Candida albicans* with DIZ, MIC, and MFC values of 10 mm, 12.5 mg/mL, and 12.5 mg/mL, respectively. The three tested *Candida* species were inhibited by the methanolic extract in a concentration-dependent manner. Indeed, the methanolic extract was found to be more potent against *Candida albicans* with higher DIZ value (12 mm) and lower MIC value (3.25 mg/mL).

Antifungal activity of extracts against the mycotoxigenic strains

In vitro evaluation of plant extracts antifungal activity by “dual culture” against *A. fumigatus*, *A. niger*, and *A. flavus* was performed. Fungal growth diameter of the tested molds decreased in a concentration-dependent manner when treated with the aqueous and methanolic extracts of *E. alata* aerial parts.

As shown in Fig. 1, fungal growth diameter of *A. fumigatus* decreased significantly ($p < 0.05$) during incubation at 25 °C for 5 days, when treated with the three highest tested concentrations of the aqueous extract and the two highest tested concentrations of the methanolic extract. However, there was no significant difference between fungal diameter of control (inoculation only with *A. fumigatus*) and fungal

diameter of those treated with the other concentrations of both extracts (Fig. 1A and B).

Indeed, fungal growth diameter of *A. niger* decreased significantly ($p < 0.05$) during incubation at 25 °C for 5 days, when treated with all the tested concentrations of both extracts and with only the two highest tested concentrations of the methanolic extract of *E. alata*. However, there was no significant difference between fungal diameter of control (inoculation only with *A. niger*) and fungal diameter of those treated with the other concentrations of the methanolic extract (Fig. 1C and D).

Also, fungal growth diameter of *A. flavus* decreased significantly ($p < 0.05$) during incubation at 25 °C for 5 days, when treated with all the tested concentrations of the aqueous and methanolic extract of *E. alata* compared with the control (inoculation with only *A. flavus*).

Indeed, the different tested concentrations of extracts exhibited various degrees of growth inhibition effects ranging from 2.23% to 18.82%, from 1.17% to 31.76%, and from 6.66% to 41.25% for *A. fumigatus*, *A. niger*, and *A. flavus*, respectively. Moreover, *A. niger* was the fungus most sensitive to both extracts, whereas *A. flavus* was the fungus most sensitive to the methanolic extract. As shown in Fig. 2, the highest fungal inhibition percentage was obtained when treated by the highest concentration of aqueous extract (Fig. 2A) against *A. fumigatus* (18.82%). Additionally, a moderate inhibition growth percentage (31.76%) was obtained with the highest tested methanolic extract followed by the highest tested aqueous extract (27.05%) against *A. niger*. *A. flavus* was more resistant to the aqueous extract (6.66–15%), whereas the methanolic extract showed a strong antagonism effect (12.5–41.25%).

Overall, our results revealed that both extracts were able to inhibit fungal growth in a concentration-dependent manner (Fig. 3).

Allelopathic activity

The allelopathic effects of the aqueous and methanolic extracts from aerial parts of *E. alata* on the germination seeds of *Triticum aestivum*, *Linum usitatissimum*, and *Trigonella foenum-graecum* were evaluated at three different concentrations, and the results are presented in

Table 1 Anticandidal activity of *Ephedra alata* extracts

Test organisms	Aqueous extract			Methanolic extract		
	DIZ ± SD	MIC	MFC	DIZ ± SD	MIC	MFC
<i>Candida krusei</i> ATCC 6258	NA	NA	NA	11 ± 0	6.25	6.25
<i>Candida neoformans</i> ATCC 14118	NA	NA	NA	11 ± 0.33	6.25	6.25
<i>Candida albicans</i> ATCC 90028	10 ± 0	12.5	12.5	12 ± 0	3.25	6.25

DIZ diameter inhibition zone, SD standard deviation, MIC minimal inhibitory concentration, MFC minimal fungicidal concentration, NA not active

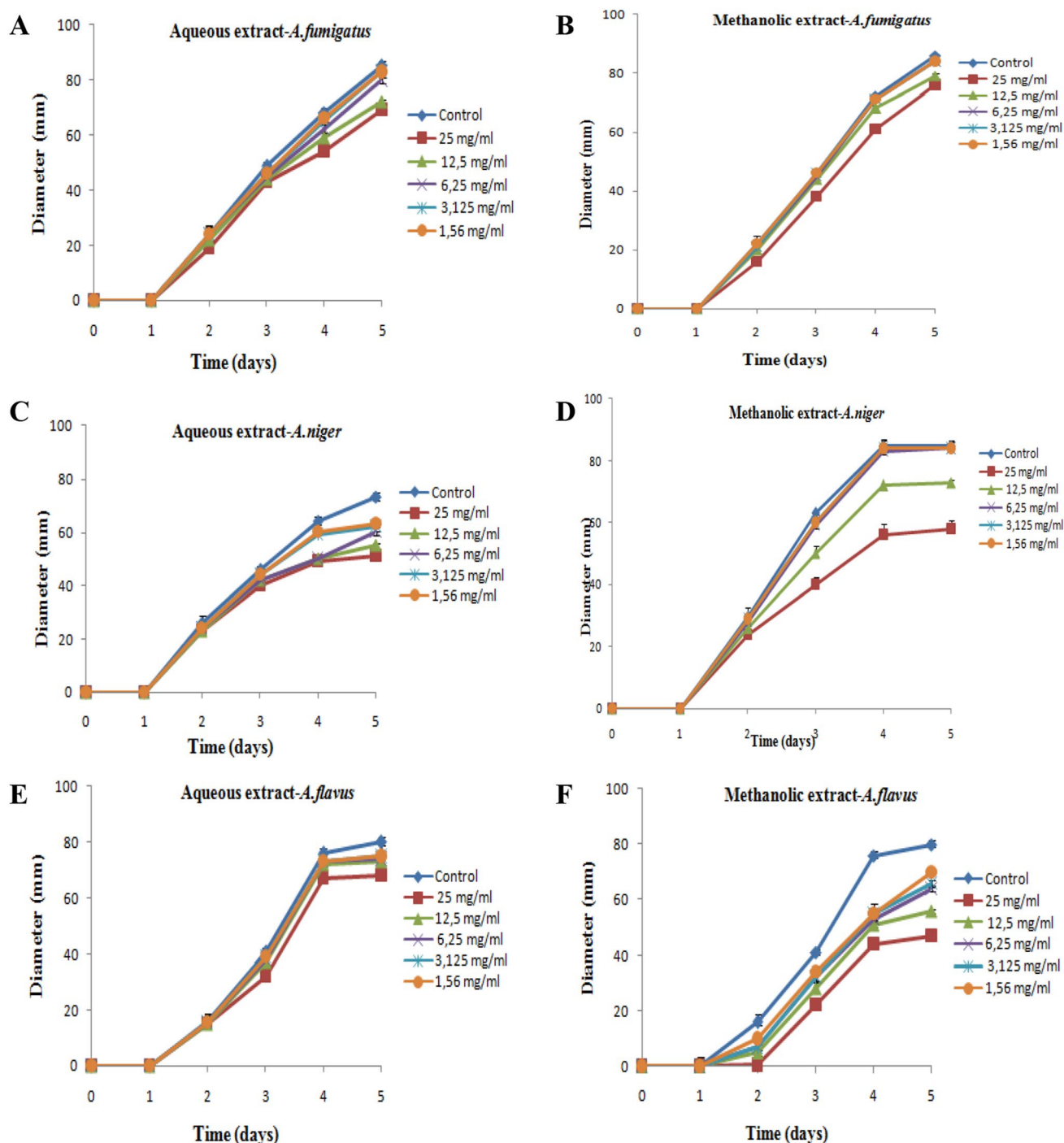


Fig. 1 Graphical illustration of *A. fumigatus*, *A. niger*, and *A. flavus* diameter co-cultured with: (A, C and E) aqueous extract and (B, D and F) methanolic extract, in comparison with control samples (plates

inoculated only with *A. fumigatus*, *A. niger*, and *A. flavus*) during incubation for 5 days at 25 °C

Table 2. Both extracts showed inhibitory activity on the germination of all the tested seeds in a concentration-dependent manner. *Linum usitatissimum* seems to have the highest sensitivity to the two extracts. For instance,

the germination inhibition percentage values of linen were 95%, 80%, and 70% for the methanolic extract, while they were 80%, 60%, and 50% for the aqueous extract at concentrations of 1%, 0.5%, and 0.25%, respectively.

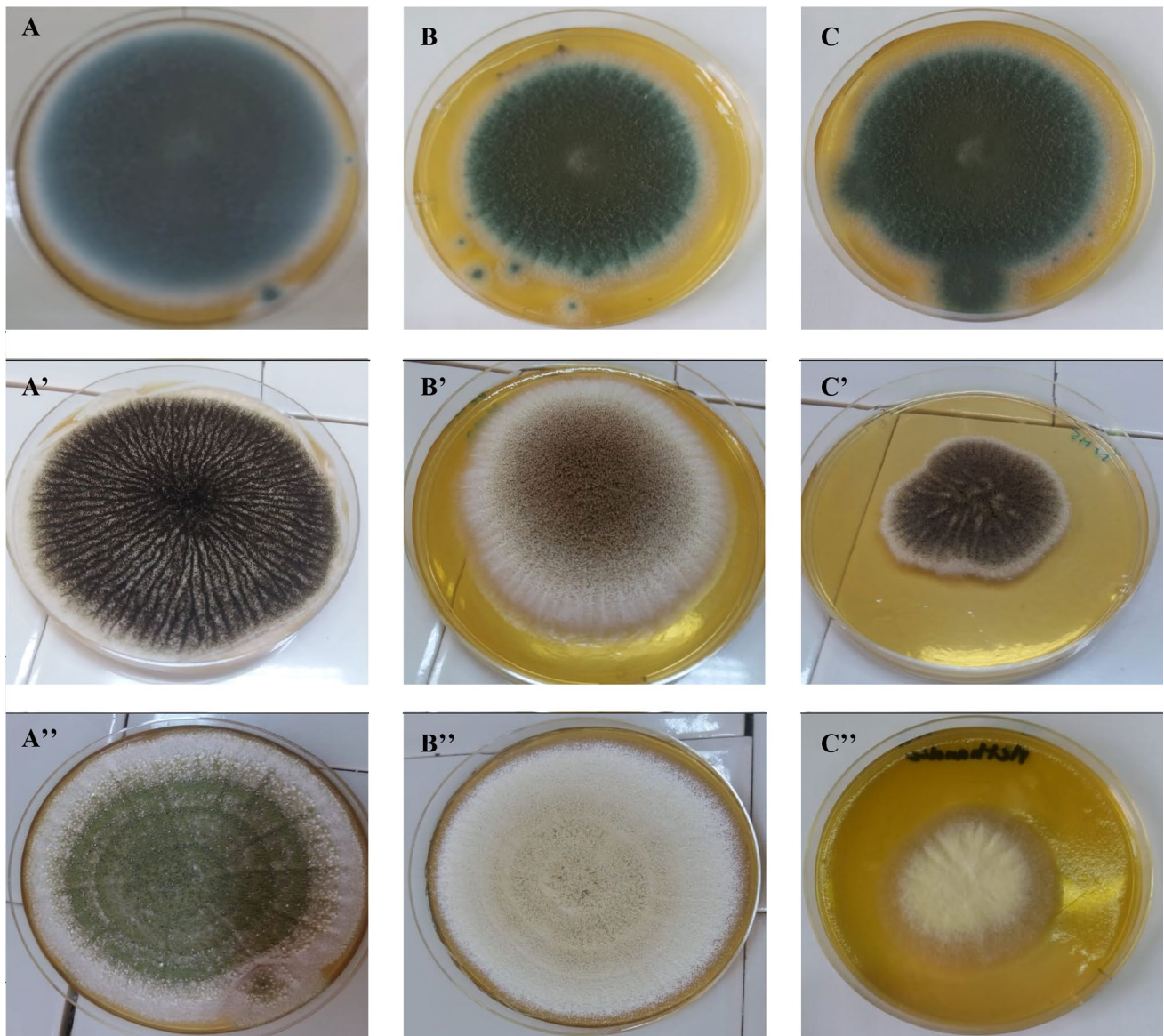


Fig. 2 Visual observation of inhibitory zone of *A. fumigatus*, *A. niger*, and *A. flavus* due to addition of the high tested concentrations (25 mg/mL) of extracts (**B**, **B'**, and **B''**: aqueous extract; **C**,

C', and **C''**: methanolic extract) in comparison with control samples (plates inoculated only with *A. fumigatus* (**A**), *A. niger* (**A'**), and *A. flavus* (**A''**) during incubation at 25 °C for 5 days

Discussion

Food and feed spoilage has become a hotspot worldwide, as it accounts for the deterioration of about 30% of total food crops products (Saladino et al. 2016). Fungal contamination and mycotoxin production are the principal cause of the deterioration of food and agricultural products (Schnürer and Magnusson 2005).

Thus, the search for biological methods that can be an effective alternative to chemical and physical strategies is needed, to fight against mycotoxigenic molds (da Cunha et al. 2018).

In this current study, aqueous and methanolic extracts of *Ephedra alata* aerial parts were tested for their potential to inhibit fungal infections associated with *Candida* and *Aspergillus* species.

Thus, we tested the anticandidal activity of the extracts against three yeasts (*Candida albicans*, *Candida krusei*, and *Candida neoformans*) that are principal causal agents of opportunistic oral, nosocomial, and genital infections in humans. The results demonstrated that the aqueous extract inhibited the growth of only *Candida albicans*, while the methanolic extract was active against all the *Candida* strains. Similarly, Danciu et al. (2019) evaluated the anticandidal activity of the hydroethanolic extract from *E. alata*, showing

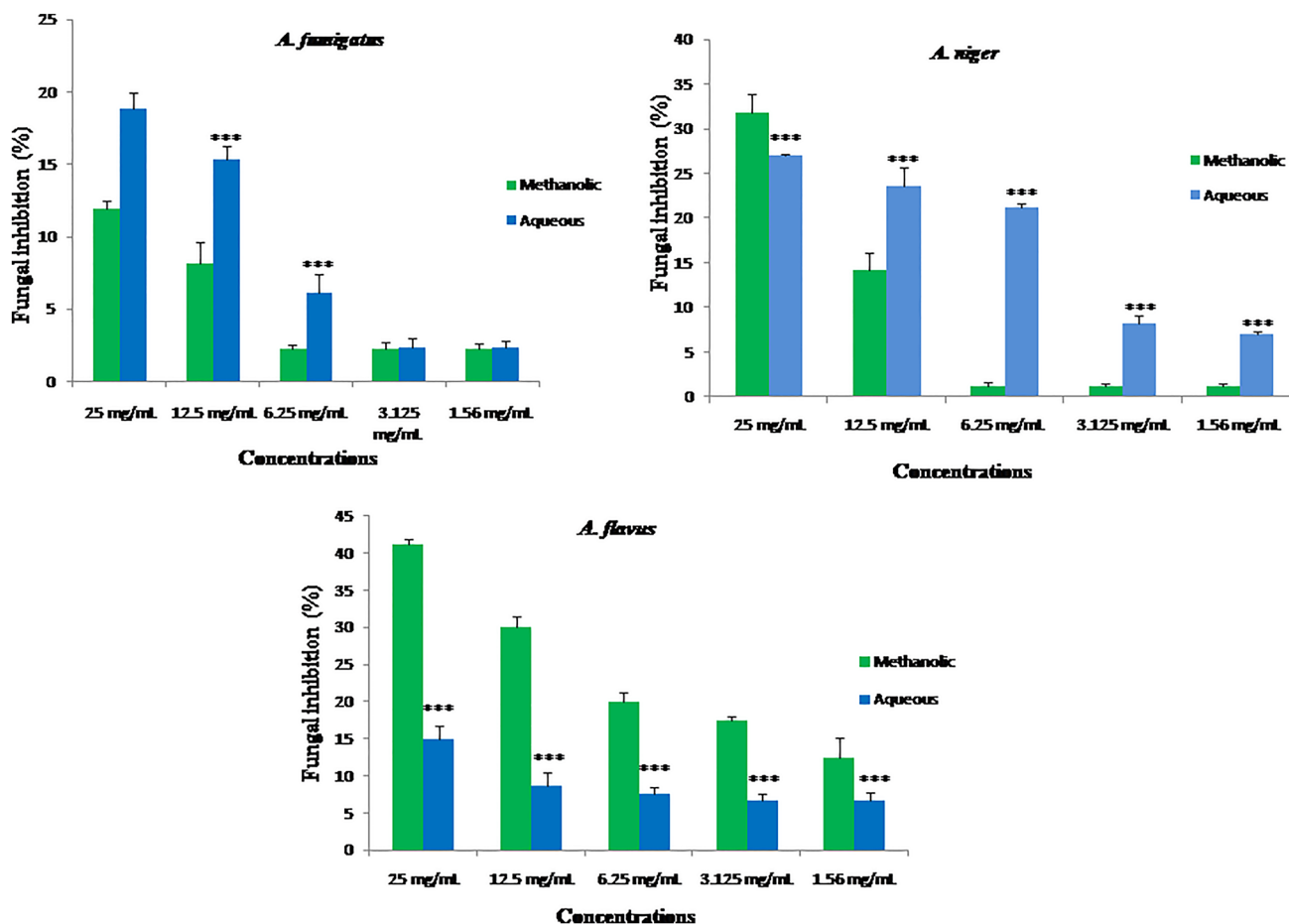


Fig. 3 Percentages of growth inhibition of *A. fumigatus*, *A. niger*, and *A. flavus* by aqueous and methanolic extracts of *E. alata* aerial parts after incubation for 5 days at 25 °C. The bars represent the

mean \pm SD. Asterisks indicate significant differences between aqueous and methanolic extracts (ANOVA, $p < 0.001$)

Table 2 Percentage germination Inhibition of wheat, linen, and fenugreek in the presence of aqueous and methanolic extracts of *Ephedra alata*

Concentrations	Germination inhibition (%)		
	<i>Triticum aestivum</i>	<i>Linum usitatissimum</i>	<i>Trigonella foenum-graecum</i>
Aqueous extract			
Control	5.00 \pm 2.64 ^a	15.00 \pm 1.00 ^f	0.00 \pm 0.00 ^{b,c}
1%	15.00 \pm 2.17 ^c	80.00 \pm 2.78 ^g	10.00 \pm 1.64 ^{b,d}
0.5%	10.00 \pm 0.60 ^b	60.00 \pm 1.74 ^h	5.00 \pm 1.00 ^{b,e}
0.25%	10.00 \pm 1.56 ^b	50.00 \pm 2.55 ^h	5.00 \pm 1.05 ^{b,e}
Methanolic extract			
Control	5.00 \pm 3.48 ^d	15.00 \pm 1.86 ^{a,b}	0.00 \pm 0.00 ^{c,b}
1%	25.00 \pm 0.85 ^d	95.00 \pm 2.91 ^{a,c}	30.00 \pm 2.64 ^{c,d}
0.5%	10.00 \pm 2.24 ^e	80.00 \pm 1.60 ^{a,d}	20.00 \pm 1.12 ^{c,e}
0.25%	10.00 \pm 1.64 ^e	70.00 \pm 2.42 ^{a,d}	5.00 \pm 0.00 ^{c,e}

Values are means of three replicates \pm standard error. Data with the same letter are not significantly different at $p < 0.05$ (Tukey's test)

that this extract was active against *Candida albicans* and *Candida parapsilosis*, with low CMI and CMB concentration values.

To assess the potential of the plant in inhibiting the growth of mycotoxigenic fungi, we tested aqueous and methanolic extracts against *Aspergillus fumigatus*, *Aspergillus niger*, and *Aspergillus flavus*.

Our results indicated that both extracts can inhibit the growth of the tested fungi in a dose-dependent manner. Moreover, the aqueous extract reduced the growth of *A. fumigatus* by 18.82%, the growth of *A. niger* by 27.05%, and the growth of *A. flavus* by 15%. Meanwhile, the methanolic extract inhibited the growth of *A. fumigatus* by 11.92%, that of *A. niger* by 31.76%, and that of *A. flavus* by 41.25%.

Similarly, Al-Qarawi et al. (2013) demonstrated that the aqueous extract of *E. alata* significantly inhibited the growth and the production of aflatoxin by *A. flavus*, aflatoxigenic seedborne mold. Moreover, Ghanem and El-Magly. (2008) reported the antifungal activity of *E. alata* extracts collected from Egypt. The results showed that the

only extract that was active against all the tested fungi was the acetonitrile extract, whereas the methanolic extract elicited only antifungal potential, with *A. fumigatus* and *P. italicum* being the sensitive strains. Many scientific investigations revealed the presence of phytochemical compounds in *E. alata* such as *cis*-methanoprolin, Citronellol, and Heptadecane, which have been allocated to its antimicrobial potency (Caveney et al. 2001; Rosato et al. 2007; Bagheri-Gavkosh et al. 2009).

For *E. alata*, to the best of our knowledge, findings that indicate its phytotoxic effects are not available. Hence, it was for the first time that we systematically evaluated and demonstrated the allelopathic inhibitory effect of *E. alata* on seed germination.

The phytotoxic effect of the aerial parts of *E. alata* by water and by the methanol solvent was evaluated in this study. Herein, the allelopathic influence on *T. aestivum*, *L. usitatissimum*, and *T. foenum-graecum* germination varied significantly according to the solvent used.

Our results showed that higher allelopathic effects on different tested seeds were obtained by methanol extraction compared with aqueous extraction.

We demonstrated that the extracts obtained from the Tunisian endemic *E. alata* aerial parts with both solvents significantly delayed the seed germination of *T. aestivum*, *L. usitatissimum*, and *T. foenum-graecum*. This inhibitory effect on weed species is directly proportional to the increase in the concentration used, and this may be due to the presence of methanolic-soluble allelochemicals such as phenolic acids. Similarly, bioherbicides produced from the extracts of natural sources have shown promising potential against weeds. Several plant extract compounds possess specific inhibiting activity against weed growth but cause no detrimental injury to crops (El-Darier et al. 2014). This may be explained by the difference in sensitivity in the target enzymes or the existence of specific receptors in weeds that recognize and react with the compounds (Kaab et al. 2020). Certain plant species have the capacity to secrete different metabolites known as allelochemicals, such as alcohols, fatty acids, phenolics, flavonoids, terpenoids, and steroids, that reduce the reproduction, growth, and development of adjacent vegetation, including weed species (Soltys et al. 2013). Allelochemical bioherbicides typically have short-lived environmental persistence and low toxicity, and they often employ multiple modes of action, which reduces the risk of herbicide resistance (Hasan et al. 2021). As a result, allelochemicals serve as good candidates for the development of bioherbicides, antimicrobial agents, and growth regulators.

Indeed, the reduction in seed germination may be attributed to the reduced rate of cell division and cell elongation due to the presence of the allelochemicals (Javaid and Anjum 2006). Likewise, allelochemicals may cause photosynthesis inhibition, a decrease in chlorophyll content,

enzymatic activity inhibition, and cell membrane and cell structure disruption (Khan et al. 2016).

To the best of our knowledge, this is the first study on phytotoxic potential of *E. alata* extracts. Phenol compounds are well known as potential phytotoxins (Seal et al. 2004). In line with our findings, early study demonstrated that the tested extracts are rich in phenolic compounds (Dbeibia et al. 2022), which can be attributed to their phytotoxic potentials. These results confirm that *E. alata* could be used as a potential allelopathic substance and should be considered as a potential “eco-friendly” herbicide source.

Conclusion

This study leads us to conclude that the aqueous and methanolic extracts from *Ephedra alata* aerial parts inhibited fungal growth in a dose-dependent manner. The aqueous extract was the most effective against *A. fumigatus*, whereas the methanolic extract possesses potent antifungal activity against the *A. niger* and *A. flavus*. Additionally, both extracts revealed allelopathic potency against the examined species *Triticum aestivum*, *Linum usitatissimum*, and *Trigonella foenum-graecum*. Such findings suggest that those extracts can be used as biological “eco-friendly” herbicide. However, further studies are required to identify the bioactive compounds from aqueous and methanolic extracts that are responsible for the observed antifungal and allelopathic potencies.

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Data availability The data sets generated during and/or analyzed during the present study are available from the corresponding author on request.

Declarations

Conflict of interest The authors declare no conflict of interest.

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