INNSPUB

Journal of Biodiversity and Environmental Sciences (JBES)
ISSN: 2220-6663 (Print) 2222-3045 (Online)
Vol. 16, No. 1, p. 1-14, 2020
https://www.innspub.net

RESEARCH PAPER OPEN ACCESS

Allelopathic management of *Avena fatua* L. (Wild Oat) pernicious weed growing in *Triticum aestivum* (Wheat) L. crop fields

Kholod A Khattab[1], Salama M El-Darier[2]

[1]*Botany and Microbiology Department, Faculty of Science, Damanhour University, Damanhour, Egypt*

[2]*Botany and Microbiology Department, Faculty of Science, Alexandria University, Alexandria, Egypt*

Article published on January 30, 2020

Key words: *Fagonia arabica*, Phytotoxins, Seedling protein electrophoresis, Zygophyllaceae, Oat, Wheat

Abstract

Study investigated the effect of aerial shoot of *Fagonia arabica* aqueous extracts (ASFAAE) on germination and some growth parameters as well as seedling protein profile of *Avena fatua* and *Triticum aestivum* in pure and mixed cultures. Phytochemical screening of *F. arabica* exerted the highest value of about (41.6 and 47.5mg/g) related to fagonine and saponin respectively and the lowest value of about (9.8mg/g) related to flavonoids. At pure culture, *A. fatua* revealed gradual decrease in germination percentage (GP), coleoptile length (CL), radicle length (RL) and seedling dry weight (SDW) by increase (ASFAAE) concentrations, while *T. aestivum* attained increase in CL, RL and SDW at low concentration level (0.5%) compare to control, on contrary (GP) revealed a gradual decrease. Generally mixed culture achieved the same results but with low values compare to pure culture. Generally, RL of *A. fatua* and *T. aestivum* were more sensitive than CL in both pure and mixed cultures. The highest percentage of genomic template stability (GTS%) achieved by *T. aestivum* in pure and mixed culture of about (92%, 84% and 76%) at 0.5, 1.5 and 2.5% respectively in pure culture, while the values reduced to (76%, 68% and 60%) at 0.5, 1.5 and 2.5% correspondingly in mixed culture. Observably GTS% of *A. fatua* achieved the lowest values in both pure (60%, 56% and 48%) and mixed (52%, 48% and 44%) cultures at 0.5, 1.5 and 2.5% respectively. Interestingly, the study recommended using ASFAAE safely as a bioherbicide to control *A. fatua* in *T. aestivum* crop fields.

***Corresponding Author:** Kholod Ali Khattab ✉ Kholodkhatab26@yahoo.com

Introduction

Allelopathy is a phenomenon, whereby a plant species chemically interferes with the germination, growth or development of other plant species has been known and documented for over 2000 years. Today the term is generally accepted to cover both inhibitory and stimulatory effects of one plant on another plant (Rizvi, and Rizvi, 1992). Nowadays, allelopathy has a significant role in research involving sustainable agriculture, like biological weed control (Duke and Lydon, 1987). The current trend is to find a biological solution to minimize the perceived hazardous impacts from herbicides and insecticides in agriculture production. In this regards, the harmful impact of allelopathy can be exploited for weed control (Batish *et al*., 2007). All plants produce compounds that are phytotoxic to another plant species at some concentration levels, these chemicals responsible for the phenomenon of allelopathy are generally referred to as allelochemicals or phytotoxins (Dayan and Duke, 2014). Many such natural compounds have the potential to induce a wide array of biological effects and can provide great benefits to agriculture and weed management (Duke and Lydon, 1987).

The word weed means any wild plant that grows at an unwanted place for example in fields and interferes with the growth of cultivated plants (Rizvi *et al*., 1981). Weeds have substantially adapted characteristics (e.g. produce an abundance of seed, rapid seedling growth, quick maturation, dual modes of reproduction, environmental plasticity) that enable them to grow, flourish, invade and dominate an important part of natural and agricultural ecosystems (Singh, *et al*., 2001; Zimdahl, 2007). In agro-ecosystems, weeds compete with crop plants for resources, interfere in crop handling, reduce crop yield and deteriorate their quality, and thus result in huge financial losses (Singh, *et al*., 2001). The use of allelopathy for controlling weeds could be either through directly utilizing natural allelopathic interactions, particularly of crop plants, or by using allelochemicals as natural herbicides reducing soil pollution and diminish auto toxicity hazards (Rice, 1984; Kohli *et al*., 1998; Kohli *et al*., 2006).

Avena fatua L. (wild oat) is considered the 13th most important pernicious worldwide weed (Holm *et al*., 1977). *A. fatua* (wild oat) is an annual grass and is difficult to remove because the grains splinter before crop maturation and many of the grains are cultivated into the soil, when they are turned up near the surface. *Triticum aestivum* (wheat) yield and high seeding rates decreased exponentially when wild oat populations increased (Khan *et al*., 2007; Almaghrabi, 2012).

Fagonia arabica is a tropical herb belonging to family Zygophyllaceae and is commonly known as 'Dhamasa'. It is a green shrub of 1 to 3 feet height found on calcareous rocks distributed throughout the Mediterranean region of Africa, Afghanistan, India and Pakistan (Rizvi *et al*., 1996; Kasture *et al*., 2014). Different parts of this herb have been used to cure various ailments, namely hematological, neurological, endocrinological and inflammatory disorders (Saeed and Wahid, 2003). Also used for skin diseases, small pox and for endothermic reaction in the body. The twigs of the plant are used as remedy for snake bite and also applied externally as paste on tumours and for the swellings of neck (Kasture *et al*., 2014).

Vegetative storage proteins that accumulate in vegetative plants tissues such as leaves, stems and tubers, depending on the plant species (El-Khawas and Shehata, 2005; Abdel-Latif *et al*., 2015; Marzouk *et al*., 2017). Sodium Dodecyl Sulphate Polyacrylamide Gel Electrophoresis (SDS-PAGE) is most economical simple and extensively used biochemical technique for determination of molecular mass of protein subunits (Sher *et al.,* 2010; Vishwanath *et al*., 2011).

The aim of the current study is to management one of the most noxious weeds (wild oat) which has a great damage effect on one of the most important crops in Egypt (wheat) depending on the allelopathic effect of ASFAAE. The results exhibited that low concentration level of ASFAAE at (0.5%) stimulate all growth parameter of wheat, while reduce all growth parameters of wild oat.

Materials and methods

Collection of Specimens

Preparation of Aerial Shoot of Fagonia arabica Aqueous Extract (ASFAAE)

Aerial parts of *F. arabica* that collected from El-Dabaa were washed and dried in an electric oven at 45°C. The dried leaves were ground to a fine powder. 0.5, 1, 1.5, 2, 2.5 and 3g was transferred to labeled bottles, and then 100mL of sterile deionized distilled water were added to each bottle. The mixture was shaken and the bottles were left for 48h at refrigerator, and then filtered to get extracts of 0.5%, 1%, 1.5%, 2%, 2.5% and 3%, the control was (distilled water).

Phytochemical Screening of Fagonia arabica

Qualitative Phytochemical Screening

Aerial shoot of *F. arabica* powder extract were analyzed to determine the presence of alkaloid (Harborne, 1999), coumarins (Harborne, 1998), fagonine (Asano *et al.*, 1994), glycosides (Lewis and Smith, 1967), Saponins (Farnsworth, 1966), steroids (Harborne, 1973) and triterpenoids were detected according to Lawand and Gandhi (2013).

Quantitative Phytochemical Screening

Extraction of total phenolic acid and total flavonoids assay was conducted using a modified method of Marinova *et al.* (2005). Total phenolics were analyzed using the Folin-Ciocalteu reagent method, according to the method of Singleton *et al.* (1999). Total flavonoids were measured by a colorimetric assay (Kim *et al.*, 2003) and tannins were determined by Folin and Ciocalteu method (Sultana *et al.*, 2012).

Germination Bioassay

Petri-dish experiment was applied to investigate the potential allelopathic effects of aerial shoot of *F. arabica* aqueous extract (ASFAAE) on germination percentage (GP), coleoptile (CL) and radicle (RL) lengths and seedling dry weight (SDW) of *A. fatua* and *T. aestivum* in pure and mixed cultures. To accomplish this experiment, 20 grains of *A. fatua* and *T. aestivum* (recipient species) were arranged in 9-cm diameter Petri-dishes on two discs of whatman No.1 filter paper under normal laboratory conditions for pure culture. On other hand mixed culture prepared by arranged 10 grains of both *A. fatua* and *T. aestivum* in 9-cm diameter Petri-dishes on two discs of whatman No.1 filter paper under normal laboratory conditions. Ten ml of ASFAAE (0.5, 1, 1.5, 2, 2.5 and 3%) or distilled water as control were added daily to three replicates in a randomized complete block design. Before sowing, the grains were immersed in 2% CHLOREX for 2 minutes then rinsed four times with distilled water. Finally, the grains were soaked in aerated distilled water for 24 hours. GP, EP, HL and RL were recorded daily for successive ten days according to AOSA (1990). After ten days, the homogenous seedling were carefully collected from each treatment, washed with tap water and then by distilled water, gently blotted with filter paper, then these seedlings were dried at 65°C till constant weight to determine the dry weight.

Calculations

Germination Percentage (GP)

Germination percentage= (Number of germinated grains/total number of grains) X 100

Numbers of newly germinated grains

Numbers of newly germinated grains were calculated according to McNair *et al.* (2012).

Numbers of newly germinated grains= (Number of germinated grains on the count day - Number of germinated grains on the previous day).

Numbers of grains not germinated

Numbers of grains not germinated on all days of the experiment were calculated according to McNair *et al.* (2012). Numbers of grains not germinated = (Total number of grains tested - Number of germinated grains on the count day).

The Reduction Percentage in Hypocotyl (RHP) and Radicle (RRP) Length

Reduction percentage (RP) was calculated according to the general equation:

Reduction percentage= [(control-allelopathic)/ control] x 100

Data analysis and Computer Programs

Data were subjected to standard analysis of variance (ANOVA) using CoStat6.303 (1998-2004) statistical analysis software manufactured by CoHort Software Company.

Growth Experiment

Growth experiment was performed to test the allelopathic effect of (0.5, 1.5 and 2.5%) of aerial shoot of *F. arabica* aqueous extract (ASFAAE) with sandy clay soil on seedling protein electrophoresis as molecular marker.

The soil samples were finally sterilized at (90°C for 48 h). Twenty grains of *A. fatua* and *T. aestivum* in pure culture (recipient species) and 10 grains of *A. fatua* and *T. aestivum* in mixed culture were sown in plastic pots (16 cm in diameter) with about 1500 g of sandy clay soil. Ten ml of ASFAAE (0.5, 1.5 and 2%) and distilled water as control were added daily to three replicates in a randomized complete block design. Before sowing, the grains were immersed in 2% CHLOREX for 2 minutes then rinsed four times with distilled water. Finally, the grains were soaked in aerated distilled water for 24 hours. The experiment was performed under normal laboratory conditions (20±2° C temperature, 75±2% relative humidity, and 14/10 h/dark photoperiod). After sixteen days, the homogenous seedling were carefully collected and SL, RL and SDW were calculated, while shoot system prepared and concealed with foil paper then put in ice and used for seedling protein electrophoresis experiment.

Seedling protein electrophoresis

For assessing the allelopathic effect of the aerial shoot of *F. arabica* aqueous extract (ASFAAE) at C, 0.5, 1.5 and 2.5% concentration, on the protein content of *A. fatua* and *T. aestivum* seedling in pure and mixed culture, sodium dodecyl sulphate polyacrylamide gel electrophoresis (SDS-PAGE) was carried out according to Laemmli (1970).

Data analysis and computer programs

Genomic template stability (GTS%) was calculated as the following:

GTS% = (1-a/n) X 100

Where a: average number of polymorphic bands detected in each treated sample, (polymorphism include appearance of new bands and disappearance normal bands), n: total number of bands (Cimino, 2006; Marzouk *et al.*, 2017).

Results

Phytochemical screening of aerial shoot of Fagonia arabica

Qualitative phytochemical screening of data listed in (Table 1) showed that *F. arabica* contains fagonine and saponins with high quantity, triterpenoids with moderate quantity, while (alkaloids, coumarins and steroids) exerted the trace quantity. Quantitative phytochemical screening showed that phenolics, flavonoids and tannins (mg/g) dry weights were significantly ($p \leq 0.05$) and attained values of about 41.6, 36.7 and 25.8 respectively (Fig. 1).

Table 1. Qualitative phytochemical analysis of aerial shoot of *Fagonia arabica* powder. + Positive, ++ strongly positive, +++ extremely positive.

Component	Alkaloids	Coumarins	Fagonine	Glycosides	Saponins	Steroids	Triterpenoids
Rate	+ ve	+ ve	+++ ve	++ ve	+++ ve	+ ve	++ ve

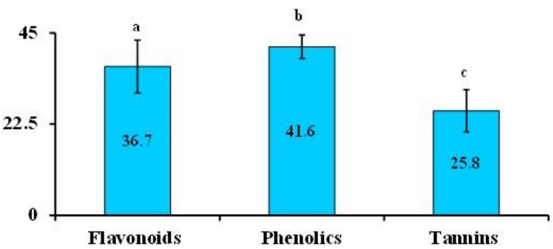


Fig. 1. Total flavonoids, phenolics and tannins (mg/g) dry weight) estimated in *Fagonia arabica* aerial shoot powder. Different letters within each column indicate a significant difference at p< 0.05 according to one way ANOVA test. Error bars indicate standard error of means.

Germination Bioassay

Germination efficiency

Bioassay experiments were carried out to test the potential allelopathic effects of aerial shoot of *F. arabica* aqueous extract (ASFAE) on number of newly added seeds, number of seed not germinated, germination percentage (GP) as well as reduction percentage in coleoptile (RCP) and radicle (RRP) lengths and seedling dry (SDW) weights of *A. fatua* and *T. aestivum* in pure and mixed culture. It was obvious that *T. aestivum* was more resistance to the effect of ASFAE compare to *A. fatua* on both pure and mixed culture.

Regression analysis was applied to predict the value of one variable (the dependent variable) on the basis of other variables (the independent variables). Exponential regression was obtained in the present study by plotting seed germination percentage (GP) of the recipient species (*A. fatua and T. aestivum* in both mixed and pure culture) versus the different concentrations of ASFAE Fig. 2. Obviously, GP decreased with increase ASFAE concentration (negative relationship) where R^2 = 0.92 in pure culture and 0.97 in mixed culture in *A. fatua* and *T. aestivum*.

Data concerning the newly added grains of *A. fatua* and *T. aestivum* in both mixed and pure cultures are illustrated and statistically represented in Fig. 3a and 3b. Petri-dish experiment demonstrated that the number of newly added seeds of *A. fatua and T. aestivum* in both mixed and pure culture was significantly ($p \leq 0.05$) affected upon applying different concentration levels of aerial shoot of *F. arabica* aqueous extract (ASFAE). Commonly, newly added seeds showed the highest value at the first day and decreased with the increase of ASFAE concentration to reach the minimum values at the last day. Generally, number of newly added seeds of *A. fatua and T. aestivum* in both mixed and pure culture decreased with the increase of ASFAE concentrations but pure culture obtained highest values compare to mixed culture. Data concerning the number of not germinated seeds of *A. fatua* and *T. aestivum* in both mixed and pure culture are illustrated and statistically represented in Fig. 4a and 4b. Petri-dish experiment demonstrated that the number of not germinated seeds of *R. sativus* was significantly ($p \leq 0.05$) affected. Generally, number of not germinated seeds of *A. fatua and T. aestivum* in both mixed and pure culture increased with the increase of ASFAE concentrations but mixed culture recorded highest values compare to pure culture.

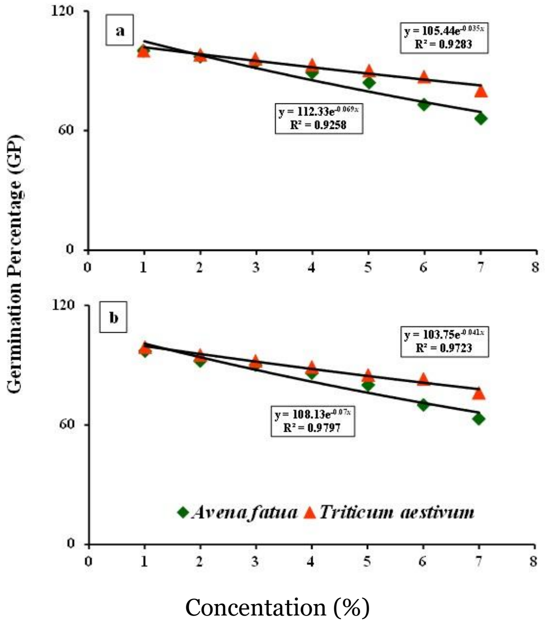


Fig. 2. Exponential regression of GP of *Avena fatua* and *Triticum aestivum* grains in (a) pure culture and (b) mixed culture (ten-days after sowing) as affected by different concentration levels (%) of aerial shoot of *Fagonia arabica* aqueous extract (ASFAE).

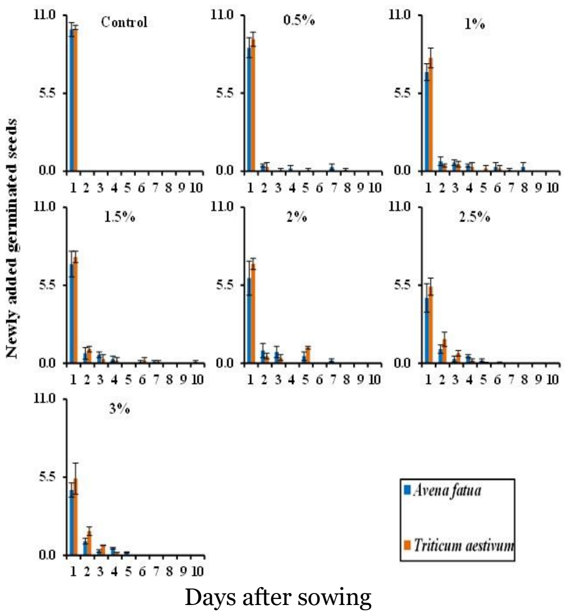


Fig. 3a. Variation in the newly added *Avena fatua* and *Triticum aestivum* germinated seeds of (ten-day days after sowing) as affected by different concentration levels (%) of aerial shoot of *Fagonia arabica* aqueous extract (ASFAE) in pure culture. Error bars indicate standard error of means.

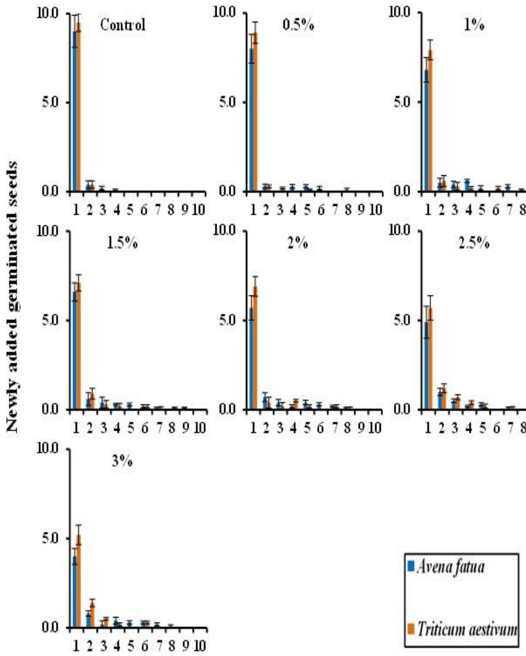
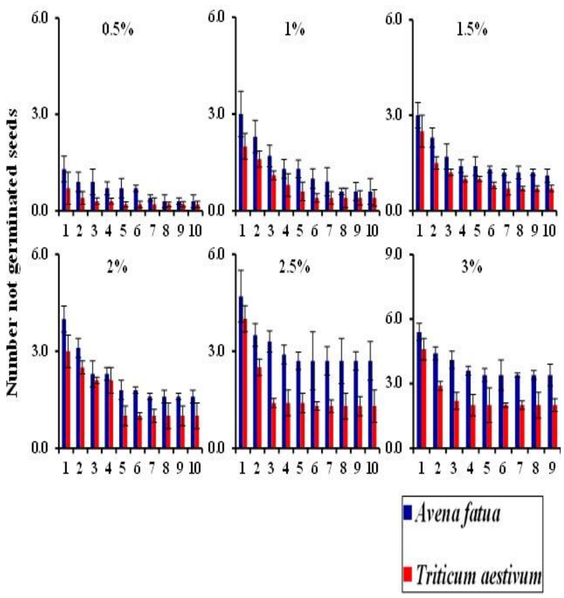
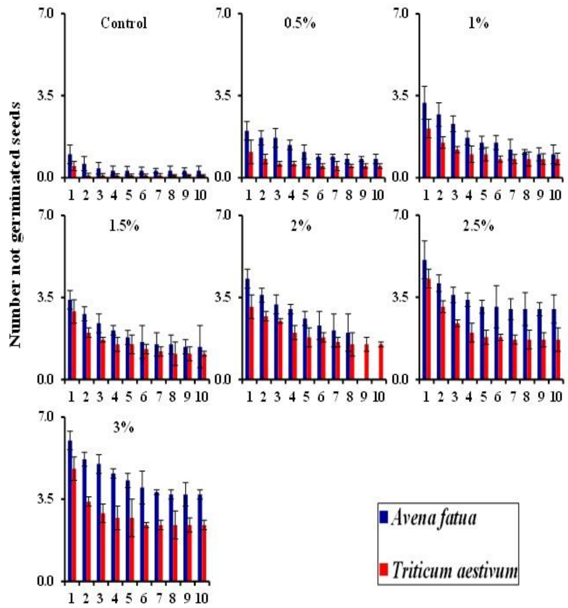


Fig. 3b. Variation in the newly added *Avena fatua* and *Triticum aestivum* germinated seeds of (ten-day days after sowing) as affected by different concentration levels (%) of aerial shoot of *Fagonia arabica* aqueous extract (ASFAE) in mixed culture. Error bars indicate standard error of means.



Days after sowing

Fig. 4a. Variation in the number of *Avena fatua* and *Triticum aestivum* not germinated seeds (ten-day days after sowing) as affected by different concentration levels (%) of aerial shoot of *Fagonia arabica* aqueous extract (ASFAE) in pure culture. Error bars indicate standard error of means.



Days after sowing

Days after sowing

Fig. 4b. Variation in the number of *Avena fatua* and *Triticum aestivum* not germinated seeds (ten-day days after sowing) as affected by different concentration levels (%) of aerial shoot of *Fagonia arabica* aqueous extract (ASFAE) in mixed culture. Error bars indicate standard error of means.

Seedling Growth Parameters

Reduction Percentage in Coleoptile (RCP) and Radicle (RRP) Lengths

The potential effect of aerial shoot of *F. arabica* aqueous extract (ASFAE) concentration on reduction percentage in coleoptile (RCP) and radicle (RRP) lengths of *A. fatua* and *T. aestivum* grains in pure and mixed cultures are illustrated and statistically represented in Fig. 5.

The corresponding allelopathic effects of ASFAE on coleoptile (CL) and radicle (RL) lengths (cm) and their reduction percentage of *A. fatua* and *T. aestivum* in pure and mixed cultures are recorded. These implications were statistically significant at ($p \leq 0.05$) as evaluated by ANOVA test Table 2.

Generally, CL and RL of *A. fatua* and *T. aestivum* in pure and mixed cultures decreased with the increase of ASFAE concentration, *T. aestivum* in pure and mixed cultures exert contrary results at 0.5% concentation level, where CL and RL recorded highest values compare to control.

Seedling Dry Weight (SDW)

The potential effect of aerial shoot of *F. arabica* aqueous extract (ASFAE) concentration on seedling dry weight (SDW) of *A. fatua* and *T. aestivum* grains in pure and mixed cultures are illustrated and statistically represented in Fig. 6. and Table 2. Seedling dry weight (SDW) of *A. fatua* and *T. aestivum* in pure and mixed cultures were greatly affected by different concentration levels of ASFAE. The effect of the applied concentrations is significant at $p \leq 0.05$ as evaluated by ANOVA test. Generally, SDW decreased with the increase of ASFAE concentration level, *T. aestivum* in pure and mixed cultures exert contrary results at 0.5% concentation level, where (SDW) recorded highest values compare to control.

Table 2. Variation in the coleoptile (CL) and radicle (RL) lengths and seedling dry weight (SDW) of *Avena fatua* and *Triticum aestivum* L. seeds as affected by different concentration levels (%) of aerial shoot of *Fagonia arabica* aqueous extract (ASFAE) in pure and mixed cultures. Different letters within each column indicate a significant difference at $p< 0.05$ according to one-way ANOVA test. ± indicate standard error.

Treatments (%)	Coleoptile length (CL) (cm)				Radicle length (RL) (cm)			
	Pure culture		Mixed culture		Pure culture		Mixed culture	
	A. fatua	*T. aestivum*	*A. fatua*	*T. aestivum*	*A. fatua*	*T. aestivum*	*A. fatua*	*T. aestivum*
C	$3.52^{a}\pm0.3$	$4^{a}\pm0.5$	$3.4^{a}\pm0.5$	$3.8^{a}\pm0.7$	$2.1^{a}\pm0.3$	$2.9^{a}\pm1.5$	$1.9^{a}\pm0.7$	$2.7^{a}\pm0.8$
0.5	$3.28^{b}\pm0.7$	$4.5^{b}\pm0.4$	$3.1^{b}\pm0.4$	$4.1^{b}\pm0.4$	$1.7^{ab}\pm0.7$	$3.4^{b}\pm1.3$	$1.5^{b}\pm0.9$	$3.1^{b}\pm0.9$
1	$3.04^{c}\pm0.9$	$3.9^{bc}\pm0.3$	$2.9^{c}\pm0.3$	$3.5^{c}\pm0.5$	$1.5^{b}\pm0.9$	$2.5^{c}\pm1.4$	$1.3^{c}\pm0.5$	$2.2^{c}\pm0.7$
1.5	$2.8^{d}\pm0.7$	$3.5^{c}\pm0.6$	$2.7^{d}\pm0.6$	$3.1^{d}\pm0.3$	$1.2^{c}\pm0.7$	$2.3^{d}\pm1.5$	$1^{cd}\pm0.8$	$2^{c}\pm0.6$
2	$2.3d^{e}\pm0.5$	$3.1^{c}\pm0.7$	$2.1^{e}\pm0.7$	$2.9^{e}\pm0.9$	$0.9^{cd}\pm0.5$	$2d^{e}\pm1.7$	$0.7^{d}\pm0.6$	$1.8^{d}\pm0.8$
2.5	$2.1^{e}\pm0.4$	$2.8^{d}\pm0.8$	$1.9^{f}\pm0.8$	$2.6^{f}\pm0.4$	$0.6^{d}\pm0.4$	$1.7^{e}\pm1.4$	$0.4^{e}\pm0.4$	$1.5^{e}\pm0.9$
3	$1.72^{f}\pm0.6$	$2^{e}\pm0.6$	$1.4^{g}\pm0.6$	$1.89^{g}\pm0.5$	$0.4^{e}\pm0.2$	$1.6^{e}\pm1.2$	$0.3^{e}\pm0.3$	$1.3^{f}\pm0.4$

Treatments (%)	Seedling dry weight (SDW) (gm)			
	Pure culture		Mixed culture	
	A. fatua	*T. aestivum*	*A. fatua*	*T. aestivum*
C	$0.053^{a}\pm0.03$	$0.11^{a}\pm0.05$	$0.043^{a}\pm0.007$	$0.09^{a}\pm0.009$
0.5	$0.046^{b}\pm0.02$	$0.15^{b}\pm0.06$	$0.036^{b}\pm0.005$	$0.13^{b}\pm0.01$
1	$0.043^{c}\pm0.01$	$0.09^{c}\pm0.05$	$0.023^{c}\pm0.009$	$0.08^{c}\pm0.005$
1.5	$0.037^{d}\pm0.02$	$0.08^{c}\pm0.06$	$0.021^{d}\pm0.008$	$0.07^{c}\pm0.006$
2	$0.035^{d}\pm0.01$	$0.07^{d}\pm0.02$	$0.019^{d}\pm0.01$	$0.06^{d}\pm0.009$
2.5	$0.032^{e}\pm0.02$	$0.06^{d}\pm0.04$	$0.017^{e}\pm0.007$	$0.04^{e}\pm0.007$
3	$0.026^{f}\pm0.01$	$0.04^{e}\pm0.03$	$0.014^{e}\pm0.003$	$0.03^{f}\pm0.006$

Growth Experiment

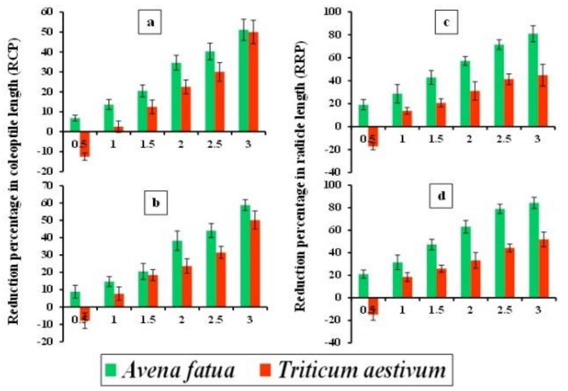
Shoot (SHL), Root (RTL) Lengths and Seedling Dry (SDW) Weight

Pot experiment was achieved to verify the effect of different levels of aerial shoot of *F. arabica* aqueous extract (ASFAE) mixed (w/w) with clay and sandy soils on some growth parameters and seedling protein electrophoresis of *A. fatua* and *T. aestivum* grains in pure and mixed cultures. A corresponding allelopathic effect of the different concentrations of ASFAE on shoot (SHL) and root (RTL) lengths and seedling dry weight (SDW) of *A. fatua* and *T. aestivum* in pure and mixed cultures was recorded in Table 3. These implications were statistically significant at ($p \leq 0.05$) as evaluated by ANOVA test. Generally, SHL, RTL and SDW decreased with the increase of ASFAE concentration level, *T. aestivum* in pure and mixed culture exert contrary results at 0.5% concentation level, where (SDW) recorded highest values compare to control.

Seedling Protein Electrophoresis

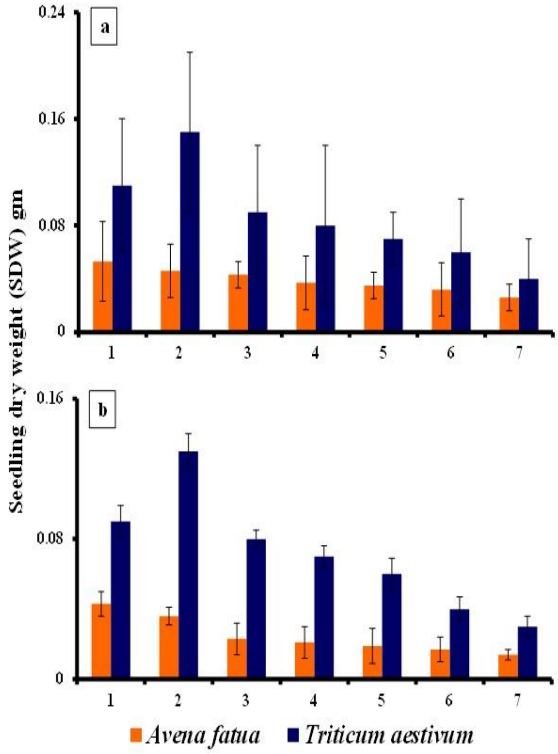
The electrograms of the *A. fatua* and *T. aestivum* specimens in pure and mixed cultures subjected to the allelopathic effects of 0.5%, 1.5% and 2.5% concentration levels of aerial shoot of *F. arabica* aqueous extract (ASFAE) relative to control level are illustrated in Plate 1. The studied specimens manifested the appearance and the disappearance of bands, as well as the alteration bands intensities Table 4. That was emphasized after calculating (GTS%) for *A. fatua* and *T. aestivum* in both pure and mixed cultures under different concentation levels. In pure culture the genomic template stability (GTS%) of *A. fatua* oscillated from 60%, 56% and 48% (subjected to 0.5%, 1.5% and 2.5% concentration level respectively).

At *T. aestivum* the values of (GTS%) fluctuated from 92%, 84% and 76% related to 0.5%, 1.5% and 2.5% concentration level correspondingly. In mixed culture the genomic template stability (GTS%) of *A. fatua* oscillated from 52%, 48% and 44% (subjected to 0.5%, 1.5% and 2.5% concentration level respectively). At *T. aestivum* the values of (GTS%) fluctuated from 76%, 68% and 60% related to 0.5%, 1.5% and 2.5% concentration level correspondingly.



Concentations (%)

Fig. 5. Variation in the reduction percentage in coleoptile (RCP) and radicle (RRP) lengths of *Avena fatua and Triticum aestivum* seeds (ten days after sowing) in pure (a and c) and mixed (b and d) cultures as affected by different concentration levels (%) of aerial shoot of *Fagonia arabica* aqueous extract (ASFAE). Error bars indicate standard error of means.



Concentations (%)

Fig. 6. Variation in the seedling dry weight (SDW) of *Avena fatua* and *Triticum aestivum* seeds in (a) pure culture and (b) mixed culture (ten-days after sowing) as affected by different concentration levels (%) of aerial shoot of *Fagonia arabica* aqueous extract (ASFAE). Error bars indicate standard error of means.

Table 3. Variation in the shoot (SHL) and root (RTL) lengths and seedling dry weight (SDW) of *Avena fatua* and *Triticum aestivum* L. seeds as affected by different concentration levels (%) of aerial shoot of *Fagonia arabica* aqueous extract (ASFAE) in pure and mixed cultures. Different letters within each column indicate a significant difference at p< 0.05 according to one-way ANOVA test. ± indicate standard error.

Treatments (%)	Shoot length (SHL) (cm)				Root length (RTL) (cm)			
	Pure culture		Mixed culture		Pure culture		Mixed culture	
	A. fatua	*T. aestivum*	*A. fatua*	*T. aestivum*	*A. fatua*	*T. aestivum*	*A. fatua*	*T. aestivum*
C	11.3^{a}±4.5	12.3^{a}±4.9	10.7^{a}±4.9	11.9^{a}±4.1	8.7^{a}±2.5	9.7^{a}±3.3	8^{a}±4.8	8.5^{a}±3.5
0.5	11^{b}±5.2	13.2^{b}±4.7	10.4^{a}±5.5	12.3b±4.3	8.2^{b}±3.4	10.2^{a}±3.5	7.6^{b}±5.2	9^{b}±3.2
1	10.5^{b} ±4.6	12.9^{c}±5.5	10.1^{b}±4.7	11^{c}±4.8	7.8^{bc}±2.9	9.4^{b}±3.8	7.2b^{c}±5.3	8.1^{c}±3.7
1.5	10^{c}±4.3	12.7^{cd}±4.8	9.8^{c}±4.5	10.7^{c}±5.2	7.5^{c}±3.2	9^{b}±2.7	6.9^{c}±4.7	7.7^{d}±3.6
2	9.7^{c}±5.5	11^{d}±4.4	9.3^{cd}±4.4	10.2^{d}±4.9	7^{c}±3.7	8.8^{c}±2.9	6.4^{c}±4.5	7.2^{cd}±2.9
2.5	9.3^{d}±4.9	10.3^{e}±5.2	8.5^{e}±4.2	9.6^{de}±4.7	6.6^{d}±3.5	8.2^{cd}±2.8	5.8^{d}±5.4	6.7^{d}±3.7
3	8.9^{de}±4.1	9.7^{f}±5.7	8^{f}±4.6	9.2^{e}±4.4	6.1^{e}±2.7	7.5^{d}±3.4	5.5^{de}±4.6	6.2^{d}±4.1

Treatments (%)	Seedling dry weight (SDW) (gm)			
	Pure culture		Mixed culture	
	A. fatua	*T. aestivum*	*A. fatua*	*T. aestivum*
C	0.19^{a}±0.04	0.35^{a}±0.08	0.16^{a}±0.02	0.29^{a}±0.05
0.5	0.17^{b}±0.06	0.41^{b}±0.09	0.15^{a}±0.04	0.32^{b}±0.06

1	$0.16^{c}±0.07$	$0.31^{c}±0.06$	$0.14^{ab}±0.05$	$0.28^{c}±0.07$
1.5	$0.15^{d}±0.08$	$0.29^{c}±0.07$	$0.13^{b}±0.03$	$0.26^{c}±0.06$
2	$0.14^{e}±0.06$	$0.25^{d}±0.05$	$0.12^{c}±0.07$	$0.23^{d}±0.08$
2.5	$0.13^{f}±0.03$	$0.22^{e}±0.04$	$0.11^{c}±0.06$	$0.2d^{e}±0.09$
3	$0.11^{f}±0.05$	$0.19^{f}±0.09$	$0.09^{e}±0.04$	$0.15^{e}±0.04$

Table 4. Changes of both total and polymorphic bands in of *A. fatua* (wild oat: O) and *T. aestivum* (Wheat: W) seeds as affected by 0.5%, 1.5% and 2.5% concentration aerial shoot of *Fagonia arabica* aqueous extract (ASFAE) in pure and mixed cultures; a, indicates appearance of new bands; b, indicates disappearance of normal bands; c, decrease in band intensities; d, increase in band intensities; a+b, denotes polymorphic bands; a+b+c+d, varied band and GTS%, genomic template stability.

Concentrations	Bands												GTS%	
	a		b		c		d		a+b		a+b+c+d			
Pure culture														
	O	W	O	W	O	W	O	W	O	W	O	W	O	W
Control	0	0	0	0	0	0	0	0	0	0	0	0	100	100
0.5%	5	1	5	1	1	2	2	2	10	2	13	6	60	92
1.5%	6	2	5	2	2	5	2	1	11	4	15	10	56	84
2.5%	7	3	6	3	1	4	3	3	13	6	17	13	48	76
Mixed culture														
	O	W	O	W	O	W	O	W	O	W	O	W	O	W
Control	0	0	0	0	0	0	0	0	0	0	0	0	100	100
0.5%	6	4	6	2	1	0	1	1	12	6	13	7	52	76
1.5%	6	4	7	4	1	2	0	1	13	8	14	11	48	68
2.5%	5	6	9	4	1	1	1	2	14	10	16	13	44	60

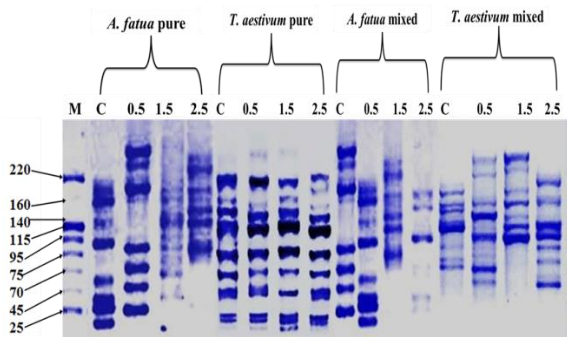


Plate 1. Seedling protein electrophoresis attained from the seedling of *Avena fatua* and *Triticum aestivum* affected by 0.5%, 1.5% and 2.5% concentration of aerial shoot of *Fagonia arabica* aqueous extract (ASFAE) in pure and mixed cultures. M indicate molecular marker and C indicate control.

Discussion

Numerous methods for weed management in different systems have been developed over the decades. Of these, herbicide, use hand and mechanical weeding are the most reliable weed control methods (Griepentrog and Dedousis, 2010; Rueda-Ayala *et al.*, 2011). Herbicides use is increasingly being adopted for weed control and for increasing crop production (Zhang, 2003). However, herbicide-resistant weeds are the major constraint to weed control in modern agriculture. Additionally, hand weeding and mechanical weed control methods are expensive and labor intensive (Gianessi, 2013). Furthermore, weed control via chemical herbicide use reportedly negatively impacts the environment and human health (Jayasumana *et al.*, 2014 and Mesnage *et al.*, 2015), that may be causes numerous health issues such as reproductive problems, neurologic impairment, pancreatic cancer, immune malfunction, and silicosis (Bhardwaj and Sharma, 2013). These challenges associated with the use of herbicides and mechanical weed control methods in land cultivation make it imperative to develop novel, environmentally friendly methods. The global interest in alternative, sustainable, and innovative technologies for weed management that not only reduce chemical herbicide use but are also environmentally and health-friendly and decrease agricultural production cost is increasing.

Jabran *et al.* (2015) improved that weeds are a hidden enemy for crop plants, interfering with their functions and suppressing their growth and development. Allelopathy as a tool can be importantly used to combat the challenges of environmental pollution and herbicide resistance development. The mechanism of action of some allelochemicals is similar to that of chemical herbicides, making them ideal for weed management. These phytochemicals are totally or partially water soluble, making them more environmentally friendly than chemical herbicides (Dayan *et al.,* 2009; Ghimire *et al.*, 2019).

In germination bioassay and growth experiments allelopathic activity of aerial shoot of *F. arabica* aqueous extracts (ASFAAE) exert a great inhibition effect relative to *A. fatua* grains and recorded the highest reduction values at high concentration level (3%) in both pure and mixed cultures that agree with Almaghrabi (2012), Cheng and Cheng (2015) and Dallali *et al.* (2017). On contrary *T. aestivum* grains revealed stimulation effect at all growth parameters in germination bioassay and growth experiments in both pure and mixed cultures at (0.5%) concentration level of (ASFAAE) and moderate reduction effect at the rest of concentration levels in concordance with El-Darier and Ahmad (2015) and Khattab *et al.* (2017). Generally (ASFAAE) has a great inhibition effect in root system compare to shoot system of both *A. fatua* and *T. aestivum* grains in pure and mixed cultures that compatible with Turk and Tawaha (2003) and Islam and Kato-Noguchi (2013).

Allelopathic activity of (ASFAAE) express their allelopathic potential by releasing allelochemicals which not only suppress the growth of *A. fatua* as noxious weed especially under high concentration levels, but also promote underground microbial activities and increase the growth parameters (0.5%) of *T. aestivum* as one of the most important crops in Egypt under small concentrations levels (Khattab *et al.*, 2017) in both pure and mixed culture. These phytochemicals are generally tannins, alkaloids, steroids, terpenes, saponins, fagonine, quinones and phenolics that recorded in aerial shoot of *F. arabica* of about 41.6 (mg/g) dry weight) that caused a great inhibition to the growth and development of *A. fatua* in both pure and mixed culture (Dayan *et al.*, 2009). Phenolic compounds consists of a hydroxyl group (-OH) bonded directly to an aromatic hydrocarbon group. In allelopathy, phenolic compounds includes simple aromatic phenols, hydroxy and substituted benzoic acids and aldehydes, hydroxy and substituted cinnamic acids, coumarins, tannins and perhaps few flavonoids (Zeng *et al.*, 2008; John and Sarada, 2012), so overwhelming evidence suggests that plant phenolics play a major role in allelopathy, that agree with Almaghrabi (2012) who found that the percentage of germination of wild oat was significantly inhibited by increasing the concentrations of phenolic compounds. At the same time, wheat and barley were slightly affected by phenolic compounds.

Almaghrabi (2012) and Radhakrishnan *et al.* (2018) found that bioherbicides based on allelochemicals especially phenolics produced by plant extracts inhibit the germination and growth of weeds by initiating damage to the cell membrane, DNA, mitosis, amylase activity and other biochemical processes and inhibit weeds seeds germination. The growth of weeds is also retarded due to low rates of root-cell division, nutrient uptake, photosynthetic pigment synthesis, and plant growth hormone synthesis, while the productions of reactive oxygen species (ROS) and stress-mediated hormones (abscisic acid and ethylene) increase, including irregular antioxidant activity.

Vegetative seedling storage protein profiling can be employed for various purposes, such as improvement and studying intercropping systems (El-Khawas and Shehata, 2005; Marzouk *et al.*, 2017; El-Darier *et al.*, 2018a), weed control and management (Abdel-Latif *et al.*, 2015; El‐Darier *et al.*, 2018b). In the present study the potential of seedling storage protein according to Laemmli method (Laemmli, 1970) to assess the genetic diversity among *A. fatua* and *T. aestivum* grains affected by 0.5, 1.5 and 2.5% concentration of aerial shoot of *F. arabica* aqueous extracts (ASFAAE) in pure and mixed culture. The results showed that most of the extracted proteins of *A. fatua* and *T. aestivum* in pure and mixed culture migrated in the range of 25 KDa to 220 KDa MW.

In the current study, the seedling protein profiles achieve relatively unstabilities as a result of allelochemicals. It is obvious through the estimation of the genomic template stability percentage, which are relatively low in comparison with the control. This is in concordance with Marzouk *et al.* (2017) and El-Darier *et al.* (2018a) found that seedling proteins decrease with the increase in the concentration of allelochemicals. Generally *T. aestivum* revealed high rate of GTS in both pure and mixed culture compare to *A. fatua* that may be due to the great inhibition effect of phenolic compounds, which reduce the incorporation of phosphorus into DNA and RNA. Meanwhile, Baziramakenga *et al.* (1997) and Padhy *et al.* (2000) concluded that phenolic acids reduced the incorporation of certain amino acid into proteins and thus reduce the rate of protein synthesis.

Conclusion and recommendations

Aerial shoot of *F. arabica* aqueous extracts (ASFAAE) can be used as a strong bio-herbicide to overcome *A. fatua* (wild oat) and can be used as a bio-fertilizer in *T. aestivum* (wheat) fields in both pure and mixed cultures due to presence of great amount of allelochemicals such in fagonine, saponins, triterpenoids, alkaloids, coumarins, phenolics, flavonoids and steroids. Phenolics have the greatest inhibition effect on *A. fatua* grains compare to *T. aestivum* grains, obviously *T. aestivum* grains exert stimulation in all growth parameters at 0.5% concentration level of (ASFAAE) compare to control in both mixed and pure cultures. Vegetative seedling storage protein profiling exert the same results whereas *T. aestivum* recorded low number of polymorphic, varied bands and high rate of genomic template stability (GTS%) near to control in both mixed and pure cultures, on contrary *A. fatua* grains recorded high number of polymorphic, varied bands and less genomic template stability (GTS%) compare to control in both mixed and pure cultures.

Obviously natural products released from allelopathic and medicinal plant residues have a great inhibition effect compare to synthetic herbicides for weed management and therefore cause less pollution and safer agricultural products (Khanh *et al.* 2007; Dallali *et al.*, 2017). The use of allelopathic potential of medicinal plants has been recommended as a viable option for alternative weed management under sustainable agriculture (Batish *et al.* 2007). Allelochemicals possess higher oxygen and nitrogen rich molecules with low heavy metals and prevent accumulation of this compound in the soil (Soltys *et al.* 2013). Allelochemicals makes them promising tools possessing specific properties to overcome weeds by binding with weed proteins at different sites than synthetic herbicides (El-Darier *et al.*, 2018b). This provides the opportunity to get rid of weeds that are already resistant to commercialized herbicides with the same mode of action. The study recommended future agricultural research will be needed to incorporate ecological, physiological and molecular methods, in order to understand agricultural crops in situ and their interaction with the environment. More advanced technological applications will be designated to explore more precise models related to allelopathy in crop agroecosystems.

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