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# Extracts obtained from organs of saffron (*Crocus sativus*) alter growth and seed germination of common lambsquarters (*Chenopodium album*) and barnyard grass (*Echinochloa crus-galli*)

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## ABSTRACT

With the aim of examining the allelopathic effects of saffron (*Crocus sativus*), *Chenopodium album* and *Echinochloa crus-galli* were selected for this study. The experiment was carried out in different levels using aqueous extract of different organs (leaf, corm and a combination of leaf and corm) with different concentrations of the extracts (0, 1, 2.5, 5, 10 and 20%). The results showed that seed germination of both weeds was significantly affected by the organs and concentration of saffron extracts. The highest germination rate of *C. album* was 2.38 seeds per day in the application of corms extract and the lowest germination rate was 2.12 seeds per day in the application of combination of leaf and corm extract. The highest seed vigor index of *C. album* was 12 in the application of leaf extract and the lowest was 9 in the application of leaf and corm extracts. In addition, the *C. album* application of the highest concentration of the extract (20%) led to the lowest weight of root, stem, seedlings, germination percentage, germination rate, seed vigor index, and number of normal seeds. In *E. crus-galli*, the increase in the concentration of the extract from 0 to 20% enhanced the mean germination time from 3.4 in control sample to 4.1 days with 20% extract concentration. All the applied extracts obtained from different saffron organs caused a significant reduction in germination traits in both species of weeds. Meanwhile the extracts of leaf and corm exerted the highest effects. Generally, the results showed that the application of 20% extract obtained from the combination of leaf and corm tissue provided the highest growth inhibition and lowest germination rate in both weeds.

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Allelopathy; corm; weed; seed vigor index

## Introduction

Weed, insects, pests, abiotic stresses and unbalanced plant nutrition are considered as major threats for crop production. Moreover, the growth of crops is also affected by temperature and disproportionate rain distribution which may result in significant plant growth and yield reduction (McDonald et al. 2009). Allelopathy can be useful in solving these problems.

low water requirement, the possibility of exploitation of 5–7 years at one planting time, long shelf life, ease of transport, no need for heavy and sophisticated machinery, the ability to employ the labor force during the harvest season, the need for irrigation during non-critical times for other plants, and the increase of saffron consumption in food, industry, and pharmaceuticals have all led to the enhancement and rapid development of saffron cultivation (Feizi et al. 2018a).

Saffron is a perennial crop with a longevity of up to 12 years, but the stands start self-thinning after five or six years, which is when the economic yield starts to decrease. For re-planting saffron, many farmers remove the top 30-cm

deep soil layer of the old saffron fields and substitute it with new soil, but this method is expensive. Various methods have been proposed for removing and/or inhibiting allelopathic effects, but most of them are impractical because of their high costs or subsequent secondary pollution. Corms and leaves of saffron (*Crocus sativus* L.) produce active compounds like safranal and crocin, which can affect other plants as well. Some of these compounds inhibit the growth and germination of the seeds of close-by plants. For this reason, many saffron growers believe that after the corms of saffron have been removed from the land, no other plants should be farmed in that land for several years (Hassani and Khalajzadeh 2011). Many researchers have elaborated the allelopathic role of saffron on the growth of other plants. Kheirabadi et al. (2020) demonstrated the aqueous extract of saffron corm remnant reduced the root length of lettuce by 50%. Feizi et al. (2018b) showed that saffron leaf and corm extracts had negative impact on sugar beet seedling weight, seedling length, and radicle length, by 4, 17 and 36%, respectively. Asgarpour et al. (2015) suggested that the application of leaf

and corm extracts of this plant results in reduced germination parameters in *Amaranthus* sp., *Sisymbrium irio* L. and *Lepidium draba* L. (*Cardaria draba* (L.) Desv.) In another study, Salari et al. (2018) showed that the negative effect of saffron leaf, corm and combination of leaf-corm extracts was significant on germination percentage, germination rate, mean germination time and weight and length of root of tomato and cumin plants. Hosseini and Feizi (2018) stated that, as saffron cultivation period increased, compounds like crocin and total phenolic increased significantly in the soil, and especially around the saffron root. Considering such content and the necessity of using natural materials to cope with damages caused by weeds, this research has been conducted on the allelopathic potential of different parts of saffron to study germination and its related parameters on common lambsquarters (*Chenopodium album* L.) and barnyard grass (*Echinochloa crus-galli* (L.) P.Beauv.).

## Materials and methods

### Plant material

Seeds of common lambsquarters and barnyard grass were collected from the farms of Agriculture College, University of Torbat Heydarieh Iran, in the summer of 2016. They were kept at room temperature until the next spring. Mean temperature was  $15 \pm 2^\circ\text{C}$ , with 20% humidity, under 8h light/16h dark regime. The duration of the light period was based on the daily natural light. To break their dormancy, seeds were treated with 98% sulphuric acid for 3 min and were thoroughly washed with distilled water for several times.

### Extract preparation

In order to obtain the extracts, the corm and leaves were separated, and in order to avoid chemical alterations, the organs were dried in greenhouse conditions for one month at  $20^\circ\text{C}$ . Afterwards, the organs were powdered using an electronic grinder and then passed through a 2mm sieve. Then 10g of the powdered organs along with 5g of the combination of each organ (5g of leaf and 5g of corm) were separately added to 100ml distilled water. Afterwards, the mixtures were put on a shaker at 200rpm for 48h. Next, the suspension solution was filtered by a first grade Whatman filter paper. These extracts have been considered as 100% stock solutions. All other tested concentrations were made by diluting the stock solution with distilled water (Asgarpour et al. 2015).

### Experimental design

This experiment was performed at the College of Agriculture and Natural Resources of the University of Torbat Haydarieh, Iran as a factorial layout based on completely randomized design with four replications. Treatments include three types of saffron extract (leaves, corms and combination of leaves and corms) in six concentrations (0, 1, 2.5, 5, 10 and 20%). Twenty-five seeds were placed in each Petri dish containing filter paper soaked with the aforementioned solutions.

According to the type of treatment, 3 ml of each extract was added to Petri dishes. Afterwards, the Petri dishes were placed inside a germinator (model TG 600) made by NST LAB Company, Iran (<https://noorsanattajhiz.com/>). The light regime was 10h light and 14h dark at temperature of  $25^\circ\text{C}$  for 14 days. During the experiment Petri dishes were watered regularly to optimize and maintain moisture conditions (Asgarpour et al. 2015). Replicates of treatments were randomly arranged within the incubator. A preliminary experiment demonstrated that 14 days was a sufficient time for the germination test since no seeds germinated after that time.

### Measurement of germination traits

The germinating seeds were counted and recorded from the third day until the fourteenth day. Seeds were considered germinated when they had a radicle at least 2mm long. At the end of the experiment, traits such as root length, stem length and seedling length, weight of root, stem and seedling, germination percentage, mean germination time, seed vigor index and number of normal seeds were measured.

In order to calculate the mean germination time, equation 1 was used (Matthews and Khajeh-Hosseini 2007):

$$\text{MGT} = \frac{\sum \text{NiTi}}{\sum \text{Ni}} \quad (1)$$

In which, MGT is the mean germination time in days, N is the number of germinated seeds in each day and T is the number of days between initiation of the experiment and germination in each stage of the experiment.

Germination rate was also calculated using equation 2 (Maguire 1982):

$$\text{Rs} = \frac{\sum \text{Si}}{\text{Di}} \quad (2)$$

Where, RS is the germination speed, Si is the number of germinated seeds in each day and Di signifies the number of days passed from the initiation of the experiment.

Seed vigor index was also calculated through equation 3 (Vashisth and Nagarajan 2010):

$$\text{VI} = \text{LS} \times \text{PG} / 100 \quad (3)$$

Where, VI is vigor index, LS signifies the seedling length and PG indicates the percentage of germination on the last day of the experiment.

### Statistical analysis

Statistical analyses were performed in SAS (Ver. 9.2) software. Additionally, mean comparison was performed using Duncan's Multiple-Range Test. The graphs were also plotted by Excel software (version 2016).

## Results and discussion

Results showed that the main effects of saffron extracts obtained from different organs were significant for common

lambsquarters root length, seedling length, seed vigor index, number of normal seeds ( $p < 0.01$ ) and its stem length and germination rate ( $p < 0.05$ ). On the other hand, the interaction between concentration and organ type was only significant for root length and seedling length at 1% and stem length at 5% significance level (Table 1).

Results of variance analyses regarding barnyard grass showed that the main effects of organ type was significant for root length, seedling length and seed vigor index at 1% significance level. In addition, the main effect of concentration was significant for all traits at 1% significance level. On the other hand, results indicated that the mutual effect of concentration in the organ type was significant for root length at 5% level and seedling length at 1% level (Table 2).

Many researchers have studied the allelopathic effect of some plant residues and their production potential and strength on the inhibition of plant growth (Narwal 2004; Feizi et al. 2018a, 2018b; Mardani et al. 2019). Germination is one of the most important stages of a plant's life. In fact, if we could control the germination process of weeds through application of certain techniques, we would finally overcome the problems caused by weeds (Narwal 2004).

In the present research, regarding common lambsquarters weed, the combination of leaf and corm extracts of saffron reduced germination compared to each of them alone. However, compared to the use of corm extract, the aforementioned decrease was not statistically significant. The highest germination rate was observed with corm extract and the least germination rate resulted from the application of the mixture of corm and leaf extract. Furthermore, the results obtained from comparing simple effects of the organs indicated that the highest number of normal germinated seeds was detected in the treatment with leaf extract with an average of seven seeds. The highest number was associated with treatment of corm extract and combined corm and leaf extract with an average of six seeds (Table 3). Mardani et al. (2015) reported that the volatile bioactive compound safranal produced by *C. sativus* has a strong inhibitory activity on plant growth and development. More recently, Mardani et al. (2019) also indicated that spraying 15  $\mu\text{M}$  of safranal caused severe injury to *Trifolium pratense* L. and could lead to the death of the plants after 48 h.

The comparisons of the means of main effects of different types of saffron organs indicated that for modification of most traits of *E. crus-galli* weed, application of the combined leaf and corm extract was more effective than leaf or corm extracts alone. In addition, no statistically significant

difference was observed between the application of leaf and corm extracts. Furthermore, the results indicated that the application of the combined solution of leaf and corm extracts results in significant reductions in seed vigor index compared to the application of leaf extract and corm extract alone (Table 4).

Mardani et al. (2019) reported that the monocot weed *Lolium multiflorum* L. was resistant to spraying with safranal. A similar trend was observed with other monocot species (*Dactylis glomerata* L. and *Phleum pratense* L.). However, *P. pratense* L. showed a significant tolerance against cell membrane disruption activity of safranal. This observation is due to the structure and genetic makeup of monocots, a common phenomenon which is related to their resistance to allelochemical and herbicides.

The comparison of means of the main effects also indicated that as the concentration of applied extract increased, all the evaluated traits had a decreasing trend, except for the mean germination time. In this regard, few traits such as root weight (52% lower than control), stem weight (19% lower than control), seedling weight (40% lower than control), germination percentage (39% lower than control), germination speed (40% lower than control) and seed vigor index (61% lower than control) were associated with highest extract concentration (20%). While applying the extract at lower densities (1 and 2.5%), no difference was recorded between the control and experimental group subjects in terms of stem weight, germination percentage, seed vigor index and number of normal seeds. As the concentration of the extract increased, mean germination time was extended in which the minimum mean germination time was observed among control subjects and the highest mean germination time (35% more than control samples) was observed as a result of applying extract at 20% concentration (Table 5). Feizi et al. (2018a, 2018b) revealed that the organs and different concentrations of saffron extract had a significant effect on seedling weight, hypocotyl weight, radicle length and seedling length of sugar beet and safflower. In another study, Mousavi et al. (2018) revealed that using the saffron extracts in the concentrations of 10 and 20% significantly negatively affected the growth and development of *Hordeum murinum* L. and *Descurainia sophia* (L.) Webb ex Prantl, weeds during the germination stage.

The comparison of the means of the main effects regarding barnyard grass indicated that as the concentration increased, most traits except for the mean germination time showed a descending trend. In terms of stem length, 10 and

**Table 1.** Results of the variance analysis for *Chenopodium album* L. germination test as affected by saffron extracts obtained from different organs and applied at different concentrations.

Treatments	Df	Number of normal seeds	Seed vigor	Speed of germination	Mean germination time	Germination percentage	Dry weight of seedlings (mg)	Dry weight of stem (mg)	Dry weight of root (mg)	Seedlings length (mm)	Stem length (mm)	Root length (mm)
Corm	2	3.59**	62**	0.40*	0.20 <sup>ns</sup>	56 <sup>ns</sup>	0.003 <sup>ns</sup>	0.0001 <sup>ns</sup>	0.001 <sup>ns</sup>	**398	23*	618**
Concentration	5	10.15**	132**	2.04**	1.58**	250**	0.033**	0.002*	0.019**	511**	47**	316**
Corm $\times$ concentration	10	0.91 <sup>ns</sup>	3 <sup>ns</sup>	0.12 <sup>ns</sup>	0.05 <sup>ns</sup>	7 <sup>ns</sup>	0.001 <sup>ns</sup>	0.0004 <sup>ns</sup>	0.001 <sup>ns</sup>	69**	14*	43**
Experimental Error	54	0.69	4	0.08	0.19	21	0.001	0.001	0.0006	16	6	11
Coefficient of variation	-	12.14	19.55	13.09	13.00	17.39	11.63	20.73	15.07	10.10	14.67	14.76

Significant at the 1% significance level \*\*, Significant at the 5% significance level \*, ns is non-significant.

Table 2. Results of the variance analysis for *Echinochloa crus-galli* (L.) P.Beauv. germination test as affected by saffron extracts obtained from different organs and applied at different concentrations.

Treatment	Df	Number of normal seeds	Seed vigor	Speed of germination	Mean germination time	Germination percentage	Dry weight of seedlings (mg)	Dry weight of stem (mg)	Dry weight of root (mg)	Seedlings length (mm)	Stem length (mm)	Root length (mm)
Corm	692**	2	0.79 <sup>ns</sup>	56**	0.35 <sup>ns</sup>	0.07 <sup>ns</sup>	12 <sup>ns</sup>	0.00002 <sup>ns</sup>	0.004 <sup>ns</sup>	0.003 <sup>ns</sup>	651**	3.69 <sup>ns</sup>
Concentration	366**	5	10.13**	150**	0.939**	0.74**	75**	0.08**	0.01**8	0.028**	864**	130**
Corm × concentration	68*	10	0.37 <sup>ns</sup>	5 <sup>ns</sup>	0.07 <sup>ns</sup>	0.06 <sup>ns</sup>	4 <sup>ns</sup>	0.0007 <sup>ns</sup>	0.001 <sup>ns</sup>	0.002 <sup>ns</sup>	94**	9.66 <sup>ns</sup>
Error	29	54	0.96	6	0.13	0.17	8	0.002	0.002	0.001	34	10.03
Coefficient of variation	15.67	–	14.01	16.70	17.02	11.02	11.19	8.97	16.62	13.65	9.74	12.23

Significant at the 1% significance level \*\*, Significant at the 5% significance level \*, ns is non-significant.

20% concentrations led to a significant decrease compared to the control group. However, in terms of other traits such as stem weight, seedling weight and seed vigor index, only 2.5–20% of the densities reduced the traits compared to control group. By increasing the concentration of solution from zero to 20%, the mean germination time increased (20.5% more than control) and the highest was associated with extract with concentration of 20% (Table 6). Amini et al. (2016) reported that stigma and style of *C. sativus* had the strongest inhibitory effects on lettuce seedling growth.

Previous studies have also reported an allelopathic property of saffron on the growth of weed and crops and the results are consistent with the present study. For example, Asgarpour et al. (2015) discussed the effect of aqueous extract of saffron on germination and growth of three types of weeds *Amaranthus retroflexus* L.; *Lepidium draba* L. (*Cardaria draba* (L.) Desv.); *Descurainia sophia* (L.) Webb ex Prantl.).

The combined extract of leaves and corms had a stronger inhibitory effect than the extracts of corm and leaves separately, on the growth of weed seeds and seedlings. Furthermore, the effect of the organ type for germination parameters was not similar between the two mentioned types of weed. The use of combined extract of saffron leaves and corm resulted in significant reduction on lambsquarter traits such as root length, stem length and seedling length, percentage and speed of germination, seed vigor index and the number of normal seeds. However, in the case of barnyard grass, the effect of organ type was not significant. It probably resulted from the difference between monocots and dicots (Mardani et al. 2019). Alipoor and Mahmoodi (2015) reported that the highest allelopathic effects of saffron corm and leaf extract on the growth and germination of *Bromus tectorum* L. are associated with leaf extract and it was associated with corm extract for *Descurainia sophia* (L.) Webb ex Prantl, It has also been reported that the more residues of saffron leaves are added to the substrate cultivation environment, the more the length of sorghum stems decrease and the more remains of saffron corm are added to the culture medium the more the length of wheat plant roots decrease. On the other hand, in this study, we observed the increasing effects resulted from adding corm remains and in some cases from adding leaf remains, which is an indication of hormetic effects (Carvalho et al. 2020). It seems that these conflicting results are due to the difference in the experimental conditions.

Moreover, in this research, the highest allelopathic effect was associated with the combined leaf and corm extract. This indicates that there are different inhibitory compounds in saffron leaves and corm, and their simultaneous presence can have the highest negative effect on growth and germination of weeds.

The comparison of the average interaction effects of the organ by concentration regarding the common lambsquarters weed has revealed that combined leaf and corm extract have reduced the root's length. The application of leaf extract did not cause any reduction in root length in any of the applied concentrations. However, the application of

**Table 3.** A comparison of the effects of extracts obtained from different saffron organs on the germination of *Chenopodium album* L. seeds.

Organ	Number of normal seeds	Seed vigor	Speed of germination (seed/day)	Mean germination time (day)	Germination percentage	Dry weight of seedlings (mg)	Dry weight of stem (mg)	Dry weight of root (mg)	Seedlings length (mm)	Stem length (mm)	Root length (mm)
Leaf	7 <sup>a</sup>	12 <sup>a</sup>	2.28 <sup>ab</sup>	3.29 <sup>a</sup>	28 <sup>a</sup>	0.34 <sup>a</sup>	0.16 <sup>a</sup>	0.17 <sup>a</sup>	44 <sup>a</sup>	16 <sup>b</sup>	28 <sup>a</sup>
Corm	6 <sup>b</sup>	10 <sup>b</sup>	2.38 <sup>a</sup>	3.38 <sup>a</sup>	26 <sup>ab</sup>	0.32 <sup>a</sup>	0.16 <sup>a</sup>	0.16 <sup>a</sup>	40 <sup>b</sup>	17 <sup>a</sup>	22 <sup>b</sup>
Combination of leaf and corm	6 <sup>b</sup>	9 <sup>c</sup>	2.12 <sup>b</sup>	3.48 <sup>a</sup>	25 <sup>b</sup>	0.31 <sup>a</sup>	0.15 <sup>a</sup>	0.15 <sup>a</sup>	36 <sup>c</sup>	17 <sup>a</sup>	18 <sup>c</sup>

\*The means that have common letters in each column have no statistically significant difference according to Duncan test at 5% level.

**Table 4.** A comparison of the effects of extracts obtained from different saffron organs on the germination of *Echinochloa crus-galli* (L.) P.Beauv seeds.

Organ	Seed vigor	Speed of germination	Mean germination time	Germination percentage	Dry weight of seedlings (mg)	Dry weight of stem (mg)	Dry weight of root (mg)	Seedlings length of (mm)	Stem length (mm)	Root length (mm)
Leaf	16 <sup>a</sup>	2.25 <sup>a</sup>	3.70 <sup>a</sup>	26 <sup>a</sup>	0.61 <sup>a</sup>	0.31 <sup>a</sup>	0.30 <sup>a</sup>	64 <sup>a</sup>	26 <sup>a</sup>	38.83 <sup>a</sup>
Corm	15 <sup>a</sup>	2.02 <sup>b</sup>	3.79 <sup>a</sup>	24 <sup>a</sup>	60 <sup>a</sup>	0.31 <sup>a</sup>	0.30 <sup>a</sup>	61 <sup>a</sup>	25 <sup>a</sup>	36.57 <sup>a</sup>
Combination of leaf and corm	13 <sup>b</sup>	2.08 <sup>ab</sup>	3.81 <sup>a</sup>	24 <sup>a</sup>	60 <sup>a</sup>	0.33 <sup>a</sup>	0.28 <sup>a</sup>	54 <sup>b</sup>	25 <sup>a</sup>	28.61 <sup>b</sup>

\*The means that have common letters in each column have no statistically significant difference according to Duncan test at 5% level.

**Table 5.** Effects of different concentrations of extract obtained from saffron organs on germination of *Chenopodium album* L. seeds.

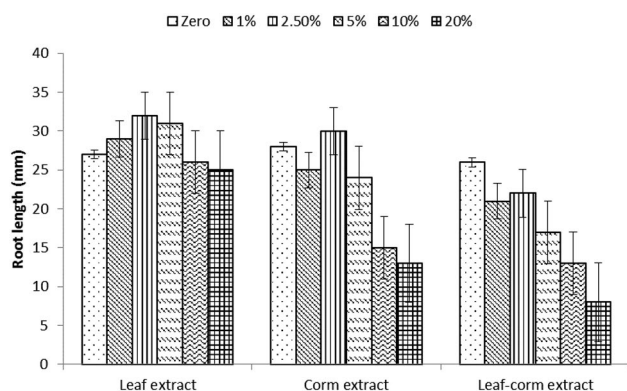
Concentration of saffron extract %	Seed vigor	Speed of germination	Mean germination time	Germination percentage	Dry weight of seedlings (mg)	Dry weight of stem (mg)	Dry weight of root (mg)	Seedlings length (mm)	Stem length (mm)	Root length (mm)
0	13 <sup>a</sup>	2.94 <sup>a</sup>	2.82 <sup>d</sup>	31 <sup>a</sup>	0.40 <sup>a</sup>	0.16 <sup>ab</sup>	0.23 <sup>a</sup>	43 <sup>b</sup>	16 <sup>bc</sup>	27 <sup>ab</sup>
1	13 <sup>a</sup>	2.48 <sup>b</sup>	3.14 <sup>c</sup>	29 <sup>ab</sup>	0.36 <sup>b</sup>	0.17 <sup>a</sup>	0.18 <sup>b</sup>	44 <sup>ab</sup>	19 <sup>a</sup>	25 <sup>b</sup>
2.5	13 <sup>a</sup>	2.25 <sup>bc</sup>	3.35 <sup>bc</sup>	29 <sup>ab</sup>	0.33 <sup>bc</sup>	0.17 <sup>a</sup>	0.16 <sup>bc</sup>	47 <sup>a</sup>	19 <sup>a</sup>	28 <sup>a</sup>
5	10 <sup>b</sup>	2.13 <sup>cd</sup>	3.52 <sup>ab</sup>	26 <sup>bc</sup>	0.30 <sup>c</sup>	0.16 <sup>ab</sup>	0.15 <sup>c</sup>	41 <sup>b</sup>	17 <sup>ab</sup>	24 <sup>b</sup>
10	7 <sup>c</sup>	2.00 <sup>d</sup>	3.67 <sup>ab</sup>	22 <sup>cd</sup>	0.30 <sup>c</sup>	0.15 <sup>ab</sup>	0.14 <sup>c</sup>	35 <sup>c</sup>	16 <sup>bc</sup>	18 <sup>c</sup>
20	5 <sup>d</sup>	1.75 <sup>e</sup>	3.81 <sup>a</sup>	19 <sup>d</sup>	0.24 <sup>d</sup>	0.13 <sup>b</sup>	0.11 <sup>d</sup>	29 <sup>d</sup>	14 <sup>c</sup>	15 <sup>c</sup>

\*The means with common letters in each column have no statistically significant difference according to Duncan test at 5% level.

**Table 6.** Effects of different concentrations of extract obtained from saffron organs on germination of *Echinochloa crus-galli* (L.) P.Beauv seeds.

Concentration of saffron extract %	Seed vigor	Speed of germination	Mean germination time	Germination percentage	Dry weight of seedlings (mg)	Dry weight of stem (mg)	Dry weight of root (mg)	Seedlings length (mm)	Stem length (mm)	Root length (mm)
0	19 <sup>a</sup>	2.09 <sup>bc</sup>	3.40 <sup>c</sup>	27 <sup>a</sup>	0.69 <sup>a</sup>	0.37 <sup>a</sup>	0.32 <sup>ab</sup>	72 <sup>a</sup>	31 <sup>a</sup>	42 <sup>a</sup>
1	17 <sup>ab</sup>	2.49 <sup>a</sup>	3.56 <sup>bc</sup>	27 <sup>a</sup>	0.68 <sup>a</sup>	0.33 <sup>ab</sup>	0.35 <sup>a</sup>	64 <sup>b</sup>	27 <sup>b</sup>	38 <sup>ab</sup>
2.5	16 <sup>b</sup>	2.33 <sup>ab</sup>	3.80 <sup>ab</sup>	27 <sup>a</sup>	0.62 <sup>b</sup>	0.32 <sup>b</sup>	0.29 <sup>bc</sup>	56 <sup>c</sup>	27 <sup>bc</sup>	36 <sup>bc</sup>
5	14 <sup>c</sup>	2.14 <sup>bc</sup>	3.86 <sup>ab</sup>	25 <sup>bc</sup>	0.61 <sup>b</sup>	0.32 <sup>b</sup>	0.30 <sup>bc</sup>	56 <sup>c</sup>	24 <sup>cd</sup>	32 <sup>cd</sup>
10	12 <sup>cd</sup>	1.97 <sup>c</sup>	3.88 <sup>ab</sup>	23 <sup>cd</sup>	0.57 <sup>b</sup>	0.29 <sup>bc</sup>	0.28 <sup>bc</sup>	52 <sup>cd</sup>	23 <sup>d</sup>	29 <sup>d</sup>
20	10 <sup>d</sup>	1.69 <sup>d</sup>	4.10 <sup>a</sup>	21 <sup>d</sup>	0.46 <sup>c</sup>	0.25 <sup>c</sup>	0.20 <sup>d</sup>	50 <sup>d</sup>	22 <sup>d</sup>	28 <sup>d</sup>

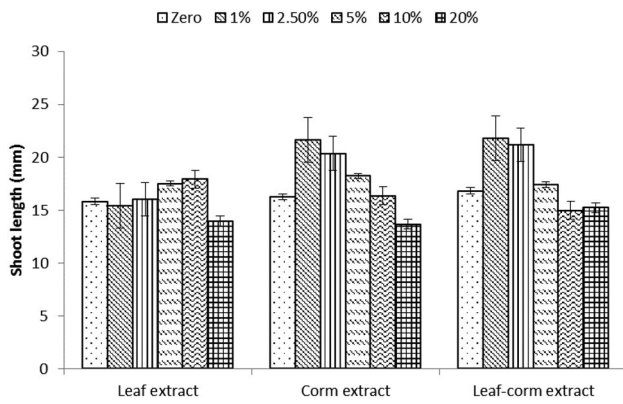
\*The means with common letters in each column have no statistically significant difference according to Duncan test at 5% level.


**Figure 1.** The significance of the interactive effect of organ  $\times$  concentration of saffron extracts on the root length in *Chenopodium album* L. germination test. Means marked with the same letters do not differ significantly at ( $P < 0.05$ ; Duncan's Multiple-Range Test).

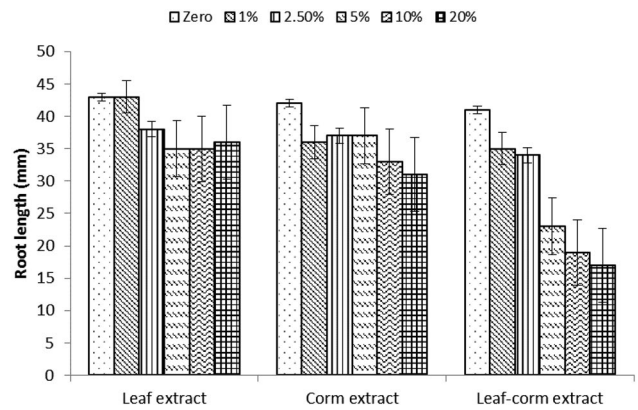
10% and 20% extracts of corm and also 5, 10 and 20% combined corm and leaf extract resulted in significant reduction of these traits compared to the control group (Figure 1).

Results regarding stem length indicated that, although this trait was reduced by 20% extract concentration, none of the other applied concentrations were able to significantly reduce this trait. (Figure 2).

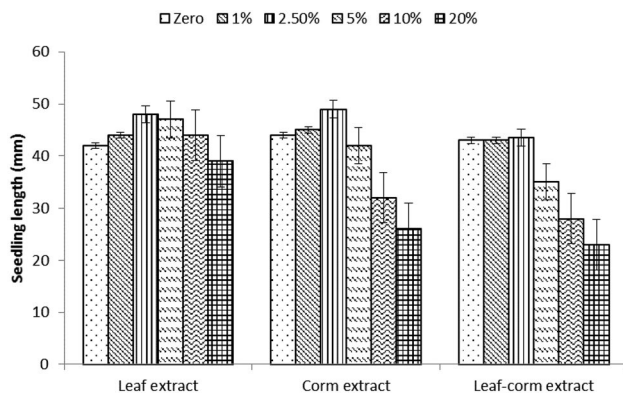
Result have also indicated that applying corm extract in 10% and 20% concentrations and the application of combined leaf and corm extract in 5%, 10% and 20% concentrations resulted in significant reduction of seedling length in lambsquarter weed. Not unlike other traits of this weed, leaf extract had a minor effect on this plant. The shortest seedling length (23 mm) was associated with the application of combined leaf and corm extract at concentration of 20%. Additionally, the highest length (49 mm) was associated with



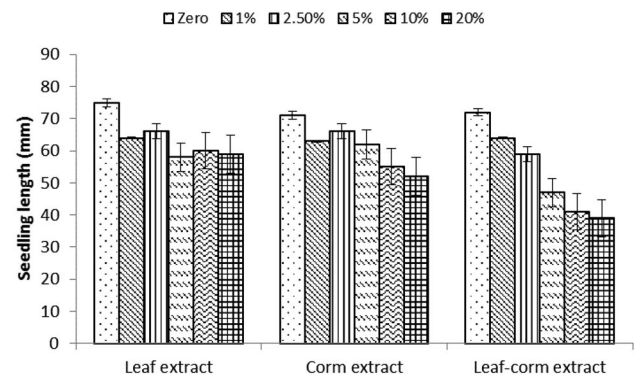
**Figure 2.** The significance of the interactive effect of organ  $\times$  concentration of saffron extracts on the stem length in *Chenopodium album* L. germination test. Means marked with the same letters do not differ significantly at ( $P < 0.05$ ; Duncan's Multiple-Range Test).



**Figure 4.** The significance of the interactive effect of organ  $\times$  concentration of saffron extracts on root length in *Echinochloa crus-galli* (L.) P.Beauv. germination test. Means marked with the same letters do not differ significantly at ( $P < 0.05$ ; Duncan's Multiple-Range Test).



**Figure 3.** The significance of the interactive effect of organ  $\times$  concentration of saffron extracts on the seedlings length in *Chenopodium album* L. germination test. Means marked with the same letters do not differ significantly at ( $P < 0.05$ ; Duncan's Multiple-Range Test).



**Figure 5.** The significance of the interactive effect of organ  $\times$  concentration of saffron extracts on the seedlings length in *Echinochloa crus-galli* (L.) P.Beauv. germination test. Means marked with the same letters do not differ significantly at ( $P < 0.05$ ; Duncan's Multiple-Range Test).

the application of corm extract at concentration of 2.5% (Figure 3). The comparison of the average mutual effects of organ by concentration for barnyard grass indicated that in all the aforementioned three extract types, as the concentration increases, the root length significantly decreases. This decreasing trend was proven to be statistically significant for the corm extract and combined corm and leaf extract. In terms of the decrease of this trait, in comparison with the control group, the highest effectiveness was respectively associated with combined corm and leaf extract and corm extract. The longest root length of barnyard grass was associated with 1% solution of leaf extract and the shortest length was recorded for 20% solution of combined leaf and corm extract (Figure 4).

All the concentrations of leaf extract and combined leaf and corm extract were able to significantly reduce seedling length in barnyard grass. Regarding the corm extract, 10% and 20% concentrations have significantly reduced the aforementioned trait compared to the control group. The longest seedling length of this weed was associated with control treatment in all the three types of extract. In addition, the shortest length was found to be associated with 20% solution of the combined leaf and corm extract (Figure 5).

In general, the results have indicated that as the concentration of saffron extract increases, germination parameters decrease in both types of weeds. In this regard, the effects of lower densities of the extract (1% and 2.5%) were not found to be significant for most traits. Even in some traits such as length of stem and seedling of the lambsquarter weed and germination speed of barnyard grass, these concentrations have led to an increase compared to the control group. Fallahi et al. (2014) indicated that allelopathic effects of saffron extract are significant in terms of entire germination indexes and seedling growth of alfalfa (*Medicago sativa* L.), arugula (*Eruca sativa* Mill.) and rapeseed (*Brassica napus* L.). In fact, as the concentration of extract increases, the percentage of germination decreases in a way that the least germination amount is associated with concentration of 6%. An almost similar trend was also observed regarding traits such as germination speed and length and dry-weight of roots and their stems. Agah et al. (2014) performed a study and indicated that as the concentration of saffron corm extract increases, a significant reduction is observed in terms of germination, root length, stem length and seed vigor in cumin plants. Their results are consistent with the results obtained from the present study. Saffron is a medicinal plant with several industrial and medicinal applications. It has been

proven that saffron extract contains flavonoid and phenolic compounds (Goli et al. 2012; Goupty et al. 2013). Among allelochemicals, aromatic compounds such as phenols, coumarone, flavonoids, tannins, derivatives of cinnamic acid and quinones are considered as the most important allelopathic substances. In addition, phenols, flavonoids, tannins and glycosides are considered as germination inhibiting compounds (Kohli et al. 2001). The mechanism which results in reduced germination is probably related to reduced activity of enzymes engaged in germination such as alpha-amylase (Vivian 2002).

## Conclusion

In general, the results of this study have shown that saffron leaf and corm extract has an allelopathic effect on the growth of *C. album* L. and *E. crus-galli* (L.) P.Beauv. during their germination period. The effects of lower concentrations of the extract such as 1% and 2.5% were not found to be significant for most traits. However, increasing the concentration to 20% caused a decline in germination-related traits in both species. In addition, the highest inhibitory effect was associated with the combined saffron leaf and corm extract at a concentration of 20%. Furthermore, the seedling growth of the weeds under study was significantly influenced by the type of saffron organ extracts. The combination of corm and leaf extract led to negative effects on the seedling length and weight of both weeds. Considering the outcome of this study, future researchers are recommended to investigate the impacts of saffron extract at other developmental stages of the species examined in order to have a better understanding of the potential of these extracts for weed control.

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## Disclosure statement

No potential conflict of interest was reported by the authors.

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