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Autotoxicity of *Amygdalus scoparia* Spach and *Atriplex canescens* (Pursh.) Nutt. on soil seed bank in Iranian arid lands

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ABSTRACT

We explored the inhibitory effects of *Amygdalus scoparia* Spach and *Atriplex canescens* (Pursh.) Nutt. organs on the soil seed bank dynamics in semi-arid areas. *Ex-situ* experiments were done using aqueous extracts [0 (control), 25, 50, 75 and 100 g.L^{-1}] of shoot, leaf and seed coat on germination indices of test grasses (*Stipa barbata*, *Bromus tectrrom*, *Hordeum leporinum*) and forbs (*Stachys inflata* and *Alyssum desertorum*). *In-situ* experiment was also done based on soil seed bank germination indices at different distances from the plant stands. Both species extracts significantly influenced the seed germination, speed of germination and vigour index. The seed coat aqueous extracts at 100 g.L^{-1} concentration was most inhibitory. Also, auto-inhibitory effects were lower than their inhibitory effects on neighboring plants species. In aqueous extracts of *A. canescens,* 4- allelochemicals identified were: Benzenecarbothioic acid, benzene, azidomethyl and 1-Hexanol, while, extracts of *A. scoparia* contained only two allelochemicals: Heptane and coumarin. The field survey showed that seeds in plant's rhizosphere had the lowest seed germination and vigour and these increased significantly with increasing distance from plants stand (R^2=0.99). The seed coat aqueous extracts of both donor species decreased the germination indices of forbs.

Key words: Allelochemicals, allelopathic effects, *Amygdalus scoparia,* aqueous extracts, *Atriplex canescens,* forbs, germination indices, grasses, secondary metabolites, seed bank

INTRODUCTION

Plant allelopathy affects the vegetation through changes in the soil seed bank dynamics (16,23). Allelopathy is defined as the negative effects of metabolic secretions of plants/microorganisms on other plants/microorganisms (1,50). In fact, the allelopathic effects caused by the secondary metabolites (5,31) may be negative (6,29,31) and include any direct/indirect effects (22). Allelopathy is an interference mechanism against neighbouring plants (44). The allelopathic effects caused by the chemicals released into the environment (15,21) affects the germination, establishment and growth of plants in natural environments (26,28,44). These chemicals (secondary metabolites) are released through leaching from the foliage, root exudates, volatiles and decomposition of plants residues (14). However, the most negative effect of allelopathy is to prevent or delay the seed germination in early stages of plant growth. Seed germination is primary, fragile and very important stage of plant life (48,51) and may be severely affected by allelopathic effects (12,44).

Allelopathic effects also significantly reduces the plant growth, hence, receiving increasing attention in natural and sustainable agricultural ecosystems (5,11,46,47). These effects depend on the host plants biochemicals, resistance of neighbouring species and environmental conditions (45,49). Thus, understanding the mechanisms of allelopathic

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effects plays an important role to develop monocultures of native and non-native species in harsh ecosystems with deficient resources. In arid and semi-arid areas, allelopathic effects are more important to develop dense monocultures. Allelopathic effects of plants are known (2,26,31,48), but little information is available on the allelopathic effects of monoculture tree species on soil seed bank dynamics in semi-arid conditions.

Both donor tree species of *Amygdalus scoparia* Spach and *Atriplex canescens* (Pursh.) Nutt. have been planted in many rehabilitation projects in degraded areas (Fig. 1).

Figure 1. A general view of *A. scoparia* (A) and *A. canescens* (B) species in study sites.

Although both these species produce large quantity of seeds, no dense and monotonous vegetation was observed in areas underneath their canopies after 17-years. We do not know the natural mechanisms of these two tree species on their soil seed bank in semi-arid lands. Therefore, this research aimed to study the allelopathic effects of *A. scoparia* and *A. canescens* trees on seeds germination indices of soil seed bank to select the proper distance between these trees in tree plantation. We studied the (i) Evidence for allelopathic effects of two donor species on seeds in rhizosphere, (ii) if there are allelopathic effects, how the inhibitory effects on seeds is affected by the distances from the plant stands in forests in semi-arid lands, (iii) allelopathic effects of aerial organs of shoot, leaf and seed coat extracts on seed germination, vigour index and speed of germination of grasses and forbs in soil and (iv) auto-allelopathy effects of plants materials on seed germination and vigour index in *A. canescens* and *A. scoparia*.

MATERIAL AND METHODS

Study sites

The studies were done in July 2019 on 17-years old two trees (*Amygdalus scoparia, Atriplex canescens)* monoculture forest trees planted at 10 x12 m spacing, in semi-arid grasslands used for livestock grazing. Therefore, after the seed dispersal from the trees, transfer and burial of seeds are mainly done by sheep and goats. The details of both sites are as under:

(i) Amygdalus scoparia: *A. scoparia* (wild almond) Rosaceae family is native medicinal plant, hence, widely distributed in Iran (18,32). It is resistant to heat and drought, hence, used in rehabilitation tree plantation projects. Its study site was Tiran rangeland central Iran (486472.00 m E; 3626880.00 m N- 487511.00 m E; 3627363.00 m N), Altitude: 2250 m asl, Annual rainfall: 252 mm, mean maximum and minimum temp were 32.5 °C and 18.6 °C, respectively, during study period.

(ii). Atriplex canescens**:** It was introduced from North and West America and first planted in Iran in rehabilitated rangelands in 1966 (19). Its study site was Akhtar-Abad rangeland (475466.00 m E; 3951772.00 m N- 476712.00 m E; 3950517.00 m N), Altitude: 1225 m asl, Annual rainfall: 255 mm, mean maximum and minimum temp were 35.4 °C and 19.3 °C, respectively, during the study period.

Vegetation and soil characteristics

Vegetation covers of 17-years old trees (density 80 ha^{-1}) of *A. canescens* and *A. scoparia* were 28.0 and 35.0 %, respectively. In these study sites, annual and perennial grasses/forbs were *Allium* spp, *Stachys inflate*, *Alyssum desertorum, Astragalus* spp, *Bromus* spp. Soil samples were collected using random sampling method and pH, EC, N, OM, P, K, $CaCO_3$ and soil texture were determined (Table 1).

Plants allelochemicals components and soil seed bank dynamics

The allelopathic effects of two donor tree species on soil seed bank indices were studied under natural field conditions and also in laboratory bioassays. Soil seed bank survey was done 2-months after the seed dispersal from the trees and burial (late July-2019). At first, a simple experiment was done to determine the seed germination in a distance gradient from stands of two species. For this purpose, soil seed bank sampling was done along a liner distance from plant stands toward bare patches. Sampling was done at distances of 0 (plant understory), 2, 4, 6, 8, 10 m away from trees *A. canescens* and *A. scoparia* stand at each site. Seed samples were collected from 50 x 50 cm plot, up to 5 cm depth (39). Seeds found at the soil surface and rotten seeds were separated from other seeds. Samples were dried and passed through 2 mm mesh sieve. The seeds of both plants were separated and their germination indices were determined.

Table 1**.** Physico-chemical properties of soil at two study sites: Tiran and Akhtar Abad

Factors	Plantation type	
	A. canescens	*A. scoparia*
Gravel {>2 cm}	11.60±3.60^{b}	33.30±7.00^{a}
Sand (%)	64.30±15.21^{a}	56.10±14.00^{a}
Silt (%)	19.50±6.50^{a}	14.25±5.85^{b}
Clay (%)	16.10±6.66^{b}	29.30±6.80^{a}
OM (%)	1.82±0.12^{a}	1.96±0.06^{a}
K (Meq/L)	75.10±11.30^{b}	98.50±14.40^{a}
$CaCO_3$ (%)	7.20±2.50^{b}	13.30±5.10^{a}
EC (ds/m)	11.71±3.30^{a}	2.64±0.15^{b}
pH	8.25±0.42^{a}	7.75±0.35^{a}
N (Meq/L)	4.35±0.65^{b}	6.20±0.55^{a}
P (Meq/L)	17.66±3.80^{b}	23.60±4.65^{a}

*Significant at 5% level

The plant's aerial parts were extracted to study the allelopathic effects of both donor species allelopathic components on soil seed bank. The extracts were also prepared from the stem bark, leaf and seed coat of both donor *A. canescens* and *A. scoparia* species. Hence, leaf, shoot and seed coats were collected from the field, first dried in the shade for 10 days and were ground to powder. Due to very small leaf of *A. scoparia*, only its stem and seed coat powder was used in experiments. Powdered material was mixed with distilled water in 1:9 ratio for 72 h, the aqueous extract (10 % weight/volume) was filtered through filter paper (5).

GC/MS (Hewlett Packard HP 5890 series II; Germany) was used to detect allelochemicals components of the plant extracts. GC-MS system was equipped with FID and a DB-5MS 5 % Phenyl Methyl Silox column (30 m × 0.25 mm; film thickness 0.25 μm). Helium was used as the carrier gas (flow rate : 1 ml/min) with the oven temperature of 60 ºC for 5 min and then programmed from 60 ºC to 280 ºC at 3 ºC /min. Mass spectra were taken at 70 eV. The mass range was from m/z 40-800 amu. The temperature of both the injector and detector was set at 280 ºC; the split ratio was 50:1 and the sample injection volume was 1 µL. Allelochemicals components was measured according to GC peak areas without FID response factor correction and Retention indices was used to identify components by comparison mass spectra data and data of National Institute of Standards and Technology. The identified chemical components, i.e. ester acid salt, amide, ether, higher chain alkane, azole, cyano compound, azide, alkaloid of two species leaves are given in Table 2.

The experimental treatments of Petriplate Lab. bioassay consisted of 3 factors: (i). Donor Tree spp. 2 (*A. canescens* and *A. scoparia*), (ii). Recipient spp 3-grasses (*Stipa barbata*, *Bromus tectrrom*, *Hordeum leporinum*) and 2-forbs (*Stachys inflata* and *Alyssum desertorum*) and (iii). Aqueous extract concentrations 5 (0,25,50,75,100 %) for both species. The treatments were replicated thrice in completely randomized design.

Table 2. Main chemical components in *A. canescens* and *A. scoparia* leaves extracts

Chemical components		Area (%)	Retention time (min)
	A. canescens		
1	Benzenecarbothioic acid	7.89	2.13
2	benzene, azidomethyl	1.95	2.33
3	2-decyloxyethanol	0.17	13.37
4	Octadecane	0.33	15.23
5	Imidazole-2,4,5-d3	0.11	16.87
6	2, 2-dideuterio-4-phenyl-butanenitrile	0.13	18.65
7	Benzamide	0.80	19.20
8	4-methoxy-1,4-dihydro-2h-pyrrole	0.26	21.22
	A. scoparia		
1	Heptane	12.8	1.86
2	Chlorobenzene	2.36	4.87
3	1-Hexanol	36.4	6.2
4	3-Octanol	3.12	8.9
5	Undecane	2.08	10.45
6	Piceol	0.86	18.75
7	Eugenol	9.11	28.2
8	Coumarin	12.33	37.6
9	Methyl palmitate	8.11	64.3
10	Palmitic acid	1.39	68.4
11	Methyl stearate	2.41	76.2
12	cis-9,10-Ethoxystearic acid, methyl ester	6.1	81.2

Recipient spp.: To study the effects of *A. canescens* and *A. scoparia* extracts on soil seed bank, seeds of most dominant grasses and forbs species in study sites were selected as Recipient spp. Grasses (*Stipa barbata*, *Bromus tectrrom*, *Hordeum leporinum*) and Forbs (*Stachys inflata* and *Alyssum desertorum*) were examined. The extracts were diluted with distilled water to prepare 25, 50, 75 and 100 % concentrations. The initial extract had 100 % concentration and other concentrations were prepared by adding distilled water to the initial extract. Distilled water was used as control.

Experiments were done in petri dishes (11-cm dia) under controlled conditions in BOD Incubator (14 h light at 25 ºC and 10 h darkness at 18 ºC and 75 % RH). Seeds were washed, sterilized with 0.5 % $HgCl_2$ and 10 seeds of each spp were sown in petri dishes lined with 2-sterilized Whatman papers. Petri-dishes were irrigated with 5 mL of test aqueous extracts in the beginning and kept in BOD Incubator. Petri dishes were irrigated twice daily with extracts. Irrigation was sufficient to wet the seed surface without water accumulation in the petri dishes.

The Auto-inhibitory effects of *A. canescens* and *A. scoparia* donor species on their own seeds germination were also determined. For this, seeds of both trees were collected from the field and the effects of their aqueous extracts were determined on seed germination. The seeds of two species were cleaned and scarified to remove the physical barriers and improve the germination, before the sowing *A. scoparia* seeds woody shells were removed. *A. canescens* seed appendages were also removed from the seed with a needle. The seeds were sown in petri dishes as described earlier.

Seed germination indices

Petri-dishes were monitored daily for seed germination and seedling growth (shoot and rootlets) for measuring following germination indices:

(i). $SG = \Sigma (G/N)*100$

Where, SG : Seed germination, G : Germinated seeds, N : Total number of seeds.

(ii). $SpG = \Sigma Fi * ni / N$

Where, SpG: Speed of germination, Fi: Number of normal seeds germinated in the time i, n: Days counted from the start of experiment, N: Total number of germinated seeds.

(iii). $GT = \Sigma Dn / \Sigma n$

Where, GT: Germination time, n: Number of seeds germinated on day D, D: Number of days from the beginning of germination.

(iv). VI (Vigour Index) = Seedling Length (cm) × germination %

Statistical Analysis

The data were Statistically analyzed by ANOVA test using SPSS 17.1 statistical package software (*Sig.*≤ 0.01). Mean comparisons were done using Duncan's test. Simple comparison and a nonlinear regression also were performed to investigate the relationship between the germination indices and distance from plant stands.

RESULTS AND DISCUSSION

Auto-allelopathy

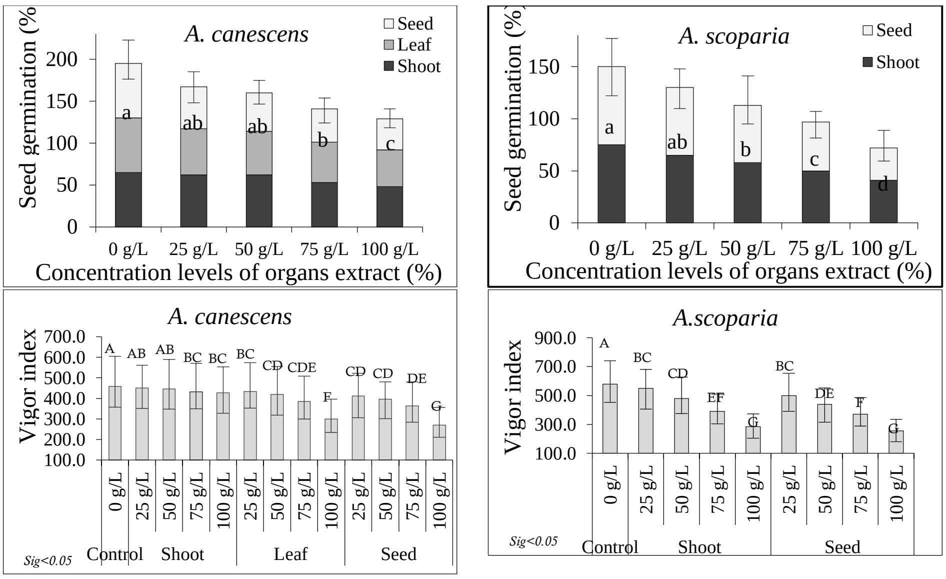

Figure 2. Auto-allelopathic effects of plants materials extract on seed germination and vigour index of *A. canescens* and *A. scoparia*.

Both donor plants of *A. canescens* and *A. scoparia* had auto-allelopathic effects on their seeds germination (Fig. 2). The auto-allelopathic effects of these two donor species were similarly inhibitory to forbs and grasses. The shoot and seed coat aqueous extracts of *A. canescens* at 100 g.L^{-1} concentration inhibited the seed germination by 26.10, 38.60 %, while, the Inhibition from *A. scoparia* seed extracts at 100 g.L^{-1} were 43.00, 55.00 % with seeds extract. The auto-allelopathic effects of *A. scoparia* were > *A. canescens*, however, the seed coat extract were more inhibitory than shoot extract (Fig. 2).

The aqueous extracts of various plant parts of donor *A. canescens* and *A. scoparia* significantly influenced the vigour index. The seed coat extracts of *A. scoparia* caused 55 % decreases in vigour index. The seeds of both donor trees had more auto-allelopathic effects on vigour index than shoot and leaf aqueous extracts (Fig. 2).

Germination and speed of germination

The plants material extracts significantly decreased the germination indices of soil seed bank ($P < 0.02$). The seed coat extracts of 75 and 100 g.l^{-1} (L3 and L4) concentrations were most harmful. All plant parts extracts influenced the germination indices in both

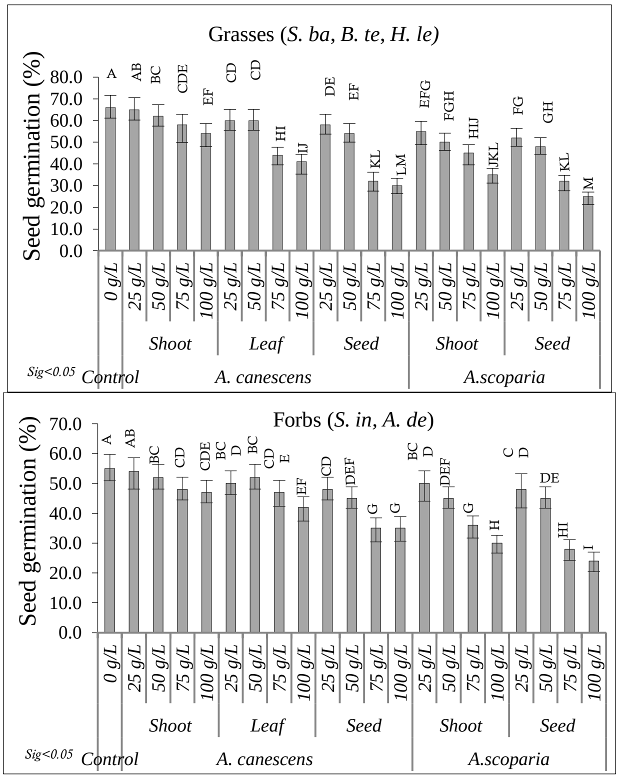

Figure 3. Effects of aqueous extracts of *A. canescens* and *A. scoparia* organs on seed germination of forbs and grasses. *(S. ba=Stipa barbata, B.te=Bromus tectrrom, H.le= Hordeum leporinum, S.in=Stachys inflate, A.de=Alyssum desertorum*)

grasses and forbs. The shoot, leaf and seed extracts of *A. canescens* reduced the seed germination by 9.0, 23.6, 36.6% in forbs and 39.4, 37.8, 54.5% in grasses, respectively. While, the shoot and seed extracts of *A. scoparia* reduced the seeds germination of forbs by 43.6, 56.3% and 39.4, 62.1% in grasses, respectively (Fig. 3). That is *A. scoparia* was more harmful to seeds germination of forbs and grasses than *A. canescens*.

Speed of germination is an important factor to determine the uniformity of seeds germination. The donor plants materials extracts had significant effects on the speed of germination (Table 3). The 100 $g.L^{-1}$ concentration of seed coat extracts significantly delayed the seeds germination of forbs. While, significant effects were not seen at 25 $g.L^{-1}$ of shoot extract. Generally, 100 % seed coat extract, caused 23 % reduction in speed of germination in grasses and forbs (Table 3).

Table 3. Effects of aqueous extracts of *A. canescens* and *A. scoparia* organs on speed of germination (days) in forbs and grasses

Plant part	Aqueous ext. conc. ($g.L^{-1}$)	*A. canescens*		*A. scoparia*	
		Grasses	Forbs	Grasses	Forbs
Control	0	4.60 ± 0.50^{C}	5.80 ± 0.40^{C}	4.60 ± 0.50^{B}	5.80 ± 0.40^{CD}
Shoot	25	4.60 ± 0.50^{C}	5.80 ± 0.60^{C}	4.70 ± 0.60^{B}	5.90 ± 0.60^{CD}
	50	4.70 ± 0.60^{BC}	5.90 ± 0.70^{BC}	4.60 ± 0.60^{B}	5.90 ± 0.60^{CD}
	75	4.80 ± 0.60^{B}	6.00 ± 0.80^{BC}	4.90 ± 0.70^{B}	6.10 ± 0.80^{CD}
	100	4.80 ± 0.60^{B}	6.00 ± 0.90^{BC}	5.10 ± 0.80^{AB}	6.10 ± 0.90^{CD}
Leaf	25	4.70 ± 0.50^{BC}	6.00 ± 0.70^{BC}	-	-
	50	4.70 ± 0.60^{BC}	6.30 ± 0.80^{ABC}	-	-
	75	4.90 ± 0.70^{B}	6.30 ± 0.90^{ABC}	-	-
	100	4.90 ± 0.70^{B}	6.50 ± 0.90^{AB}	-	-
Seed	25	4.80 ± 0.60^{B}	6.00 ± 0.60^{BC}	5.2 ± 0.7^{AB}	6.20 ± 0.80^{BCD}
	50	4.90 ± 0.70^{B}	6.60 ± 0.80^{AB}	5.4 ± 0.8^{AB}	6.50 ± 0.80^{BC}
	75	5.20 ± 0.70^{AB}	6.80 ± 0.90^{A}	5.8 ± 0.8^{A}	6.90 ± 0.90^{AB}
	100	5.80 ± 0.80^{A}	6.90 ± 1.00^{A}	6.0 ± 0.9^{A}	7.20 ± 0.90^{A}

($P < 0.05$)

Also, there was a non-liner relation between the seeds germination in soil at different distances from the donor plant stands of *A. canescens* and *A. scoparia*. Both donor trees spp. understory rhizosphere had seed germination inhibitory zone of 8-9 m from the tree base and thus, there was steep slope near plants distances in schemes (Fig. 4). The polynomial regression (with maximum R^2) showed seed germination trend in relation to distance from the plant stand. The most significant effects were concluded from the schemes based on the R^2 understory plants (=0.97-0.99) (Fig. 4).

Soil seed bank has an important role in conserving, managing and restoring lands especially in semi-arid ecosystems (18,32,41). However, successful management of ecological resources depends on the knowledge of plant connections dynamics (11) and allelopathic effects are negative and play an important role in regulating the plant community structure (27). Based on the results of this research, both *A. canescens* and *A. scoparia* showed potential inhibitory effects on the soil seed bank dynamics and shoot, leaf and seed coat had variable allelopathic effects. It was shown that *A. scoparia* had higher allelopathic effects on grasses seed germination. In this regard, previous study indicates that by increasing the extract concentration of aerial parts of *A. canescens,* the inhibitory allelopathic effects increased in *Artemisia sieberi* (24). The chemical compounds in shoots, fruits and leaves of *A. canescens* significantly influenced the seed germination and speed of germination in *Salsola rigida* (15). For instance, increase in Na^+ accumulation in *A. canescens* tissues influences the biological processes in neighboring plant species (37).

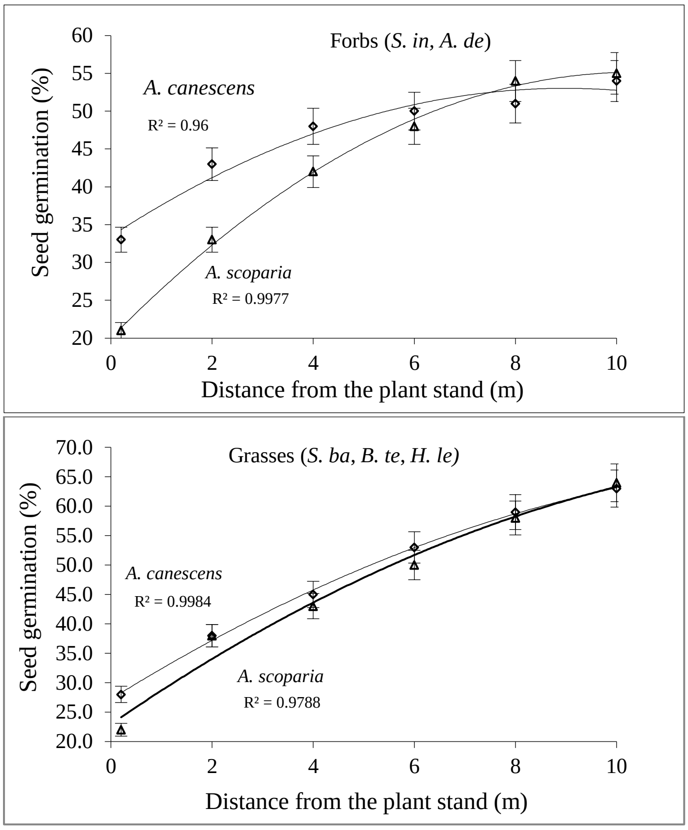

Figure 4. Seed germination in forbs and grasses in relation to distance gradient from plants stands. *(S. ba=Stipa barbata, B.te=Bromus tectrrom, H.le= Hordeum leporinum, S.in=Stachys inflate, A.de=Alyssum desertorum*)

Secondary metabolism of total tocopherols, wax and phenolics contents of *A. scoparia* seeds are high (20), which may be inhibitory to the soil seed bank. GC/MS also showed that 1-Hexanol, Heptane and Coumarin were the major components in plant organs. For example, *Cryptomeria japonica* extracts drastically inhibits the seed germination of *Robinia pseudoacacia* (43). Similarly, other studies have been reported inhibitory allelopathic effects of *Zanthoxylum limonella*, *Argemone ochroleuca, Artemisia kopetdaghensis, Mentha piperita* species that mainly inhibit seed germination of *Amaranthus tricolor*, *Farsetia aegyptia, Salvia aegyptiaca, Lactuca sativa* species (4,10,13). In general, allelopathic effects differ based on species, habitats, the organ and its concentration of extracts. In most cases, highest inhibition occurs at 100 % concentration of shoots or other plant organs extract (3,6,8,26).

Seeds vigour

The vegetative parts aqueous extracts of both donor species (*A. canescens, A. scoparia*) significantly inhibited the vigour index of test forbs and grasses (Fig. 4). The shoot extract of both trees had a similar influence on grasses and forbs. However, the seed coat aqueous extracts of *A. scoparia* seeds were most inhibitory to vigour index. Using the seed coat extracts concentration of 100 g/L of *A. scoparia*, the vigour index in grasses and forbs were 73.0 and 71.8 %, respectively. While, using 100 g/L concentration of *A. canescens* seed extracts, it was 71.0 and 57.0 in grasses and forbs, respectively (Fig. 5).

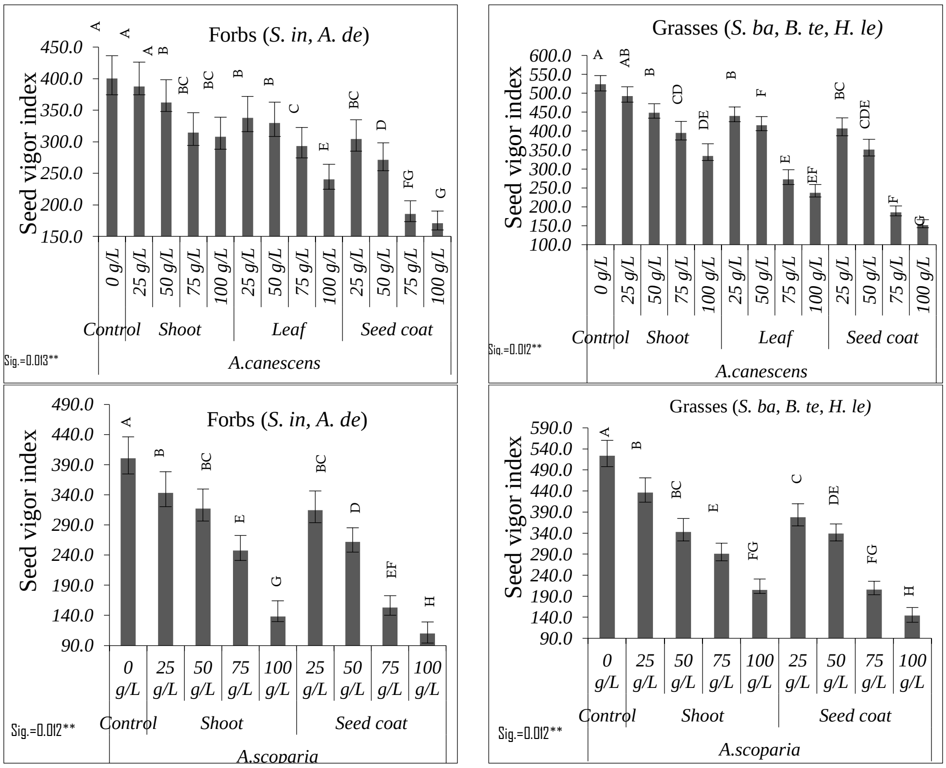

Figure 5. Seed vigour index of forbs and grasses under aqueous extracts of *A. canescens* and *A. scoparia* organs. *(S. ba=Stipa barbata, B.te=Bromus tectrrom, H.le= Hordeum leporinum, S.in=Stachys inflate, A.de=Alyssum desertorum*)

Vigour index is the most sensitive indicator of plants extracts (6) and a delay in endospermic mobilization of storage carbohydrates under plants extracts may be one of the issues to increase the speed of germination, which leads to a change in vigour index (7).

Seedling growth

The vegetative parts aqueous extracts of both species severely inhibited the seedling growth of forbs and grasses (Fig. 6). The shoot extract of *A. canescens* and *A. scoparia* had similar influence on grasses and forbs. However, the seed coat aqueous extracts of *A. scoparia* seeds were most inhibitory to seedling growth.

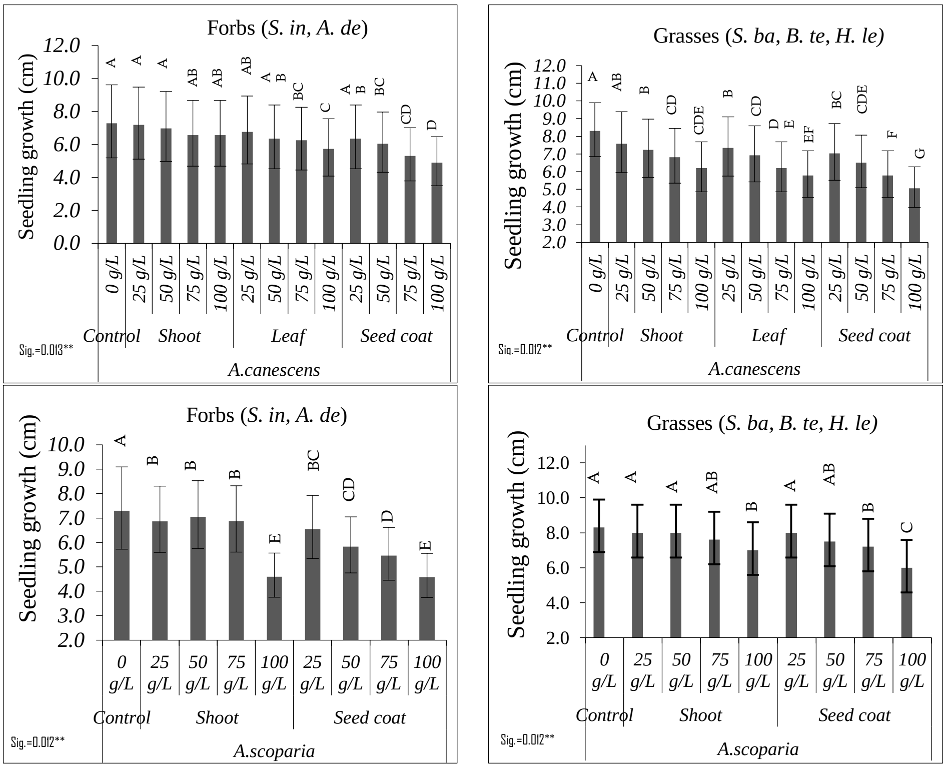

Figure 6. Seedling growth of forbs and grasses under aqueous extracts of *A. canescens* and *A. scoparia* organs. *(S. ba=Stipa barbata, B.te=Bromus tectrrom, H.le= Hordeum leporinum, S.in=Stachys inflate, A.de=Alyssum desertorum*)

Allelopathy influences the soil seed bank and seedling growth, causing negative plant-soil microbial communities interactions (30,52). These interactions are strongly influenced by climate and dry conditions make these changes more severe. The germination of grasses and forbs was significantly influenced by different concentrations of both species extracts, particularly at 100 % seed coat extract. Likewise, the *Zanthoxylum limonella* fruit extract at 2500 μM concentration completely inhibited seedling growth of *Amaranthus tricolor* L. and *Echinochloa crus-galli* (L.) Beauv. (10)

Moreover, germination indices of grasses and forbs significantly increased with increase in distance from the plant stands (Sig. < 0.002). It seems that two specie changed the micro-bio-chemical characteristic of soil and created an inhibitory environment to prevent seed germination, growth and establishment of plants. In fact, litter releases many allelochemicals in the soil that significantly inhibits the growth of plants (25) as a natural mechanism of competition for limited resources (38). Also, in both *A. canescens* and *A. scoparia* organs, especially seeds coat significantly influenced their germination indices. However, allelopathic effects depend on the concentration of inhibitory materials in plants organs. In some cases, plants leaf (4,48) strongly inhibits the seeds germination but, in other plants fruits are most inhibitory (4,35,36).

Generally, allelopathic effects are complex, which inhibits the growth and establishment of neighboring species (33,34) particularly in pure tree plantation in arid and semi-arid areas. Therefore, more knowledge is needed for plant selection and management strategies in stressful environments without or with fewer allelopathic effects (6).

CONCLUSIONS

The results of laboratory and field studies confirmed the allelopathic effects of donor *A. canescens* and *A. scoparia* trees on soil seed bank dynamics through allelochemicals present in their different organs. These trees allelopathic effects adversely affected the seedlings growth of test grasses (*Stipa barbata*, *Bromus tectrrom*, *Hordeum leporinum*) and forbs (*Stachys inflata* and *Alyssum desertorum*). Both donor trees seed coat aqueous extracts at 100 g.l^{-1} concentration were most inhibitory to seeds germination of forbs and grasses. The auto-inhibitory effects of two donor trees significantly decreased their seeds germination and vigour index. Furthermore, based the field data, the maximum seed germination was observed at 6-8 m distance away the donor tree stands.

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CONFLICT OF INTEREST

The authors announce that they have no conflict of interest.

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