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Phytochemical profile and allelopathic potential of *Haloxylon scoparium* Pomel (Chenopodiaceae) from Algerian Sahara

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Abstract: The aim of the present work is to study the chemical composition, to estimate the phenolic compounds content and to evaluate the potential allelopathic effects of the *Haloxylon scoparium* Pomel. Phytochemical tests revealed that *Haloxylon scoparium* contains tannins, saponins, coumarins, alkaloids, flavonoids and steroids. Furthermore, it contains high levels of total phenolic (588.33 mg GAE 100 g^{-1}) and flavonoids (95.45 mg QE 100 g^{-1}) contents. Moreover, LC-MS-MS analysis allowed us to determine their chemical composition. The results of this characterization confirm the presence of vanillin, naringenin, folic acid, maleic acid, benzoic acid, myricetin, qwuercetin, beta-carotene, butylhydroxyanisole (BHA), butylated hydroxytoluene (BHT), rutin, cafeic acid, hydroxy-4-coumarine, ascorbic acid, and gallic acid. The allelopathic effect was studied on seed germination and seedling growth of four weed species. The bioassays were performed using different concentrations (1 %, 2.5 %, 5 % and 10 %) against a negative control. The seed germination, shoot and root length of weed species were completely inhibited at the highest concentrations (10 %, 5 %). However, the lower concentrations exhibited lesser inhibition percentages on the germination and the seedling growth. The phytochemical results and the significant allelopathic effects of the plant extract suggest that this species may offer new substances for the biocontrol of weeds.

Key words: phytochemical profile, allelopathic potential, *Haloxylon scoparium*, LC-MS-MS analysis, allelochemicals, Algerian Sahara

Fitokemični profil in alelopatski potencial vrste *Haloxylon scoparium* Pomel (Chenopodiaceae) iz alžirske Sahare

Izvleček: Namen raziskave je preučiti kemijsko sestavo in vsebnost fenolnih spojin za ovrednotenje alelopatskega potenciala vrste *Haloxylon scoparium* Pomel. V raziskavi je bilo ugotovljeno, da vrsta vsebuje različne tanine, saponine, kumarine, flavonoide, alkaloide in steroide. Vsebuje velike količine celokupnih fenolov (588,33 mg GAE 100 g^{-1}) in flavonoidov (95,45 mgQE 100 g^{-1}). Podrobnejša kemijska LC-MS-MS analiza je pokazala prisotnost vanilina, naringenina, folne, jabolčne in benzoične kisline, mircetina, kvercetina, beta-karotena, butil hidroksianizola (BHA), butiliranega hidroksitoluena (BHT), rutina, kavne kisline, hidroksi-4-kumarina, askorbinske in galne kisline. Alelopatski učinek je bil preučevan na kalitvi in rasti kalic štirih vrst plevelov. Preizkušene so bile različne koncentracije alelopatskih snovi (1 %; 2,5 %, 5 % in 10 %) napram kontroli. Kalitev semen in dolžinska rast korenin in poganjkov plevelov je bila popolnoma zavrta pri največjih koncentracijah alelopatskih snovi (10 %, 5 %). Manjše koncentracije alelopatskih snovi so pokazale manjši odstotek zaviranja kalitve in rasti sejank plevelov. Alelopatski učinek izvlečkov te rastline nakazuje, da bi ta rastlina lahko bila vir novih učinkovin pri biokontroli plevelov.

Ključne besede: fitokemični profil, alelopatski potencial, *Haloxylon scoparium*, LC-MS-MS analiza, alelokemikalije, alžirska Sahara

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1 INTRODUCTION

Weed interference in agricultural fields reduces the quantity and quality of crops, causing enormous economic losses for farmers (Sarić-Krsmanović et al., 2019). Control strategies of weed have relied mainly on the application of synthetic herbicides. However, the continuous and excessive application of these treatments cause environmental pollution, negative effects on human health and unsafe agricultural products. Moreover, this practice increases herbicide resistance in weeds (Batish et al., 2007). Therefore, in order to resolve this problem, and minimize the dependency on chemical herbicides for weed control, great efforts have been given to develop natural and eco-friendly alternatives (Bhadoria, 2011).

Allelopathy refers to any process that involves allelochemicals produced by plants. Some plants may inhibit seed germination, emergence and growth of other plants by exuding toxic substances. These substances are called allelopathic chemicals or allelochemicals (Monem, 2012). Biochemical compounds offer a great potential for the discovery of new environmentally safe herbicides, referred to as "bioherbicides". Thus, the usage of plant secondary metabolites proved to be a promising solution in biological control (Weih et al., 2008).

The Algerian Sahara, known for its richness in spontaneous plants, harbors about 500 species of higher plants, some of which are used as medicinal plants. The *Haloxylon scoparium* Pomel plant, belonging to the Chenopodiaceae family, and locally named "*remth*", is used in traditional medicine to treat eye disorders (Chehma, 2006; Salah et al., 2002).

Several studies have been carried out on this species extract; mostly targeting its' polyphenol contents or its' biological activities. This plant has been reported to possess antidiabetic potential (Benkherara et al., 2021), antimicrobial and antiradical properties (Drioiche et al., 2019), antibacterial and antioxidant activities (Bouaziz et al., 2016), antidiabetic, antiseptic and anti-inflammatory effects (Ziyyat et al., 2014), anticancer activity (Bourogaa et al., 2014), anti-leukemic agent (Bourogaa et al., 2011), molluscicidal activity (Mezghani- Jarraya et al., 2009), anti-cancer and anti-plasmodial activities (Sathiyamoorthy et al., 1999), larvicidal activity (Sathiyamoorthy et al., 1997).

The allelopathic potential of *Haloxylon scoparium* on various weeds and crops have rarely been investigated (Salhi, 2011). Therefore, this study aims to evaluate the allelopathic effects of *Haloxylon scoparium* leaf extract on seed germination and seedlings growth. As well as to determine the chemical composition we carried out extraction of phenolic compounds, then their qualitative and quantitative characterization. The analytical techniques used are phytochemical tests based on precipitation or coloration of extract by specific reagents; Determination of total polyphenol and total flavonoids contents was determined by spectrophotometry and identification of phenolic compounds by Liquid Chromatography- Mass Spectrometry (LC-MS-MS).

2 MATERIAL AND METHODS

2.1 EXTRACT PREPARATION

The leaves of *Haloxylon scoparium* were harvested from the plant in its' natural habitat, located in the northeastern region of the Algerian Sahara, during its' vegetative stage. The leaves were subsequently washed with tap water then shade dried. The dried leaves were ground using an electric blender and stored in glass jars until the extraction process. The plant extract was obtained using reflux extraction. In a flask, 50 g of plant material were added to a hydro-methanolic solution. Using a flask heater, the mixture was heated at 60 °C for six hours. Filtration was then carried out using Whatman No. 1 filter paper. The collected filtrate underwent treatment under reduced pressure in a rotary evaporator in order to eliminate the methanol. The recovered aqueous extract was subsequently stored at 4 °C in a refrigerator until used for the biological testing (Kemassi et al., 2019).

To evaluate the dose dependent effect on the germination and seedling growth of weed, different concentrations were prepared from the stock solution, diluted with distilled water (10 %, 5 %, 2.5 % and 1 %). Distilled water served as control.

2.2 PHYTOCHEMICAL TESTS

The leaf extract was examined for the presence of the following phytochemical classes, using the numerous standard methods of evaluation described by various authors in the scientific literature

2.2.1 Tannins

A solution of $FeCl_3$ (5 %) was added to the crude extract. The presence of tannins was indicated by the appearance of a black or a blue-green color (Lerato et al., 2017).

2.2.2 Anthocyanins

Two milliliters of leaf extract, two milliliters of HCl (2 N) and ammonia (2 ml) were mixed. The appearance of a pink red coloration that turned blue violet confirmed the presence of anthocyanins (Lerato et al., 2017).

2.2.3 Saponins

Distilled water was added to the crude leaf extract in a test tube, followed by vigorous stirring. The formation of a persistent froth confirmed the presence of saponins in the extract (Lerato et al., 2017).

2.2.4 Coumarins

A NaOH solution (10 %) was added to the leaf extract. The formation of a yellow color confirmed the presence of coumarins (Lerato et al., 2017).

2.2.5 Alkaloids

The presence of alkaloids was assessed by adding three milliliters of HCl (1 %) to three milliliters of crude extract. The mixture was heated for twenty minutes. Subsequently, Mayer's reagent was added in drips to the mixture. The formation of a cream precipitate or the occurrence of a green coloration indicates the presence of alkaloids (Lerato et al., 2017).

2.2.6 Flavonoids

The plant extract was treated with a NaOH solution (10 %). The appearance of an intense yellow color of the solution indicated the presence of flavonoids (Lerato et al., 2017).

2.2.7 Steroids

Chloroform and H_2O_4 were added to the leaf extract. The presence of steroids was indicated by a color change, from violet to blue or green, or the occurrence of a blue-green ring (Lerato et al., 2017).

2.2.8 Carbohydrates

A boiled mixture of Fehling solutions A and B, with equal volumes, was added to the leaf extract. A red colored precipitate indicated the presence of reducing sugars (Jaradat et al., 2015).

2.3 DETERMINATION OF POLYPHENOLS COMPOUNDS CONTENT

2.3.1 Determination of total polyphenol content

Total polyphenol content (TPC) was measured according to the colorimetric method (Singleton and Rossi, 1965), with some modification (using a UV spectrophotometer). 200 µl of leaf extract were added to 1 ml of Folin-Ciocalteu reagent (diluted 10 times with distilled water) and 800 µl of Na_2CO_3 (7.5 %). The mixture was then incubated at 50 °C for 30 minutes. Subsequently, the absorbance of the solution was measured at 765 nm. Gallic acid served as a control for the creation of a calibration curve to estimate the TPC (Cliffe et al., 1994).

2.3.2 Determination of total flavonoid content

The total flavonoid content (TFC) was measured according to the Aluminium Chloride colorimetric method (Djeridane et al., 2006), with some modification. In a test tube, 25 µl of the plant extract was mixed with 300 µl of $NaNO_2$ and 300 µl of $AlCl_3$ (10 %) and left for five minutes. 100 µl of NaOH (2 %) were added. The absorbance of this mixture was measured at 510 nm. Calibration curve of standard quercetin solution was prepared to calculate TFC (Kim et al., 2003).

2.4 EXTRACTION OF PHENOLIC COMPOUNDS FOR LC-MS-MS ANALYSIS

The crude extract was fractionated (liquid-liquid) by the addition of ethyl acetate to obtain ethyl acetate fraction. Extract fraction was dried and dissolved in 5 ml of methanol then stored at 4 °C until analysis.

2.5 LIQUID CHROMATOGRAPHY-MASS SPECTROMETRY ANALYSIS CONDITIONS (LC-MS-MS)

The analysis of the *Haloxylon scoparium* leaf extract was carried out in the Technical Platform of Physico-Chemical Analysis (PTAPC-CRAPC)-Ouargla-Algeria, using a UPLC-ESI-MS-MS Shimadzu 8040 Ultra-High sensitivity with UFMS technology was employed and

equipped with binary bump Nexera XR LC-20AD identified the extracted phenolic compounds. For optimization of polyphones standards, we used direct injection without column. All standards were prepared in methanol with a 500 µg l^{-1} concentration. The ion trap mass spectrometer was used in both positive and negative ions with MRM mode (multiple reaction monitoring). The mobile phase was made of water, 0.1 % formic acid and 70 % methanol. The injection volume was six µl and the flow rate was 0.3 ml min^{-1}. The samples were separated using an ultra-force C18 column (I.D. 2.5 mm × 100 mm, 1.8 µm particle size; Restek), the oven temperature was 25 °C. Isocratic elution was applied with 0.1 % formic acid and methanol. The injection volume was 10 ml and the flow rate was 0.30 ml min^{-1} (Ben amor et al., 2022).

2.6 BIOASSAY EXPERIMENT

The bioassay experiments were arranged in a completely randomized design, with four replications of each treatment. Seeds of *Bromus rubens* L., *Phalaris minor* L., *Plantago lagopus* L. and *Ammi visnaga* L. were placed on filter paper in sterile Petri dishes and treated with three ml of different concentrations of plant extract. The control was treated with three ml of distilled water. The petri dishes were kept under laboratory conditions with day temperature ranging from 19-24 °C and night temperature from 12-15 °C. The germination assessment was evaluated daily, for ten days, by counting the number of germinated seeds, measuring the shoot and the root lengths and determining fresh mass at the end of the experiment (Otmani et al., 2022).

2.7 EVALUATION OF ALLELOPATHIC EFFECTS

The allelopathic effects can be defined as the inhibition or the retardation of seed germination and reduction or stimulation of root and shoot length and mass.

2.7.1 Calculation of inhibition percentage

The inhibition percentage was calculated according to the equation proposed by Côme (1970). This parameter explains the ability of a plant extract to inhibit seed germination. In the equation, mentioned below, N is the number of germinated seeds, and A is the total number of the sown seeds.

$$Inhibition\ (\%) = ((A-N)/A)*100 \quad (1)$$

2.7.2 Average germination time

The average germination time (AGT) was determined through daily counting of germinated seed to the tenth day and calculated with the equation proposed by Labouriau (1983), being the expressed results in days.

$$AGT = \Sigma ni*ti/\Sigma ni = \\ = (n_1*t_1+n_2*t_2+ \ldots +n_n*t_n)/n_1+n_2+\ldots\ldots+n_n \quad (2)$$

n_1: number of germinated seeds at time t_1.
n_2: number of germinated seeds at time t_2.
n_n: number of germinated seeds at time t_n.

2.7.3 Effects of extract on seedling's growth

After the germination test, the shoot and root lengths of the weed species were measured. Afterwards, the seedlings were separated into shoot and root parts in order to measure the fresh mass.

2.8 STATISTICAL ANALYSIS

The results obtained from the various experimental tests were analyzed by one-way ANOVA with the "XLSTAT version 2014" software. Results were evaluated by the Fisher LSD test

(p = 0.05), and presented as mean ± SD (Standard deviation).

3 RESULTS AND DISCUSSION

3.1 PHYTOCHEMICAL TESTS

The results of the phytochemical screening, presented in Table 1, clearly indicate the presence of different secondary metabolites in *Haloxylon scoparium* leaf extract. These tests revealed the presence of phenols (tannins, saponins, coumarins, flavonoids), alkaloids and steroids. However, the absence of carbohydrates, anthocyanin and betacyanin is noted.

The phytochemical tests carried out on the leaf extract of *Haloxylon scoparium* allowed us to highlight the presence of several phytochemicals. These research results are in agreement with those obtained in previous studies that indicated the richness of *Haloxylon scoparium* in secondary metabolites (Haida et al., 2020). Ben kherara et al. (2021) confirmed the presence of six major compounds (alkaloids, flavonoids, saponins tan-

Table 1: Results of phytochemical tests of *Haloxylon scoparium* Pomel. leaf extract

Constituents	Leaf extract
Tannins	+++
Anthocyanin and Betacyanin	-
Saponins	+
Coumarins	+++
Alkaloids	+++
Flavonoids	+++
Steroids	+++
Carbohydrates	-

+++: Strong positive result, ++: Moderate positive result, +: Weak positive result, - : Negative result

nins, anthocyanins, terpenes and sterols) and the absence of two other important compounds (leucoanthocyanins and cardinolids). A study done by Lachkar et al. (2021) showed that the aqueous and organic extracts of the aerial part of *Haloxylon scoparium* collected in Taza (Marroco) contains catechic tannins, flavonoids, saponins, alkaloids, anthracenosides, and free quinones. However, gallic tannins, sterols and anthraquinones were absent. Furthermore, Bourogaa and collaborators (2014) revealed the presence of flavonoids and alkaloids, while quinones and sterols are absent from the aqueous extract. In contrast, Zerriouh (2015) showed that the aqueous extract of the aerial part of *Hammada scoparia* collected in Algeria is devoid of flavonoids but contains the alkaloids and saponins.

These secondary metabolites possess allelopathic effects on the seed germination and seedling growth of weed species. These phytochemicals present in *Haloxylon scoparium* leaf extract might be controlling the observed allelopathic activity of the plant extract. alkaloids, flavonoids, terpenoids, curcurbitacins, glycosides, coumarins, saponins and tannins are the plant components identified as allelochemicals in the allelopathic effects of several plant extracts on weeds and crops (Mseddi et al., 2018; Naz and Bano, 2013).

3.2 TOTAL POLYPHENOL AND FLAVONOIDS CONTENT

The total polyphenol and total flavonoids contents obtained for leaf extract of *Haloxylon scoparium* are presented in Table 2. The total polyphenols content of the plant extract was determined in comparison to the standard gallic acid, expressed as mg GAE 100 g^{-1} of dry plant sample, whereas the total flavonoids content was measured in comparison to the standard quercetin, and expressed as mg QE 100 g^{-1} of dry plant sample.

Table 2: Quantitative results of *Haloxylon scoparium* Pomel. leaf extract

Constituents	Leaf extract
Total Polyphenols Content (mg GAE/100 g DM) ± SD	588.33 ± 1.87
Total Flavonoids Content (mg QE / 100 g DM) ± SD	95.45 ± 1.21

GAE: Gallic acid equivalent; QE: Quercetin equivalent; DM: dry mass

These interesting results of colorimetric analysis show a very high content of total polyphenols (588.33 ± 1.87 mg GAE g^{-1} DM) and total flavonoids (95.45 ± 1.21 mg QE 100 g^{-1} DM) in the *Haloxylon scoparium* leaf extract. These amounts were significantly better than those found by Allaoui et al. (2016) who obtained a high content of total polyphenol (397.743 mg GAE g^{-1} of extract) and flavonoid (82.835 mg QE g^{-1}). The obtained results were three time higher than those quantified in the same studied plant species (Zeghada, 2009; Lachekar et al., 2021; Ben kherara et al., 2021)

The extraction yields depend on the plant species, part of plant used, period of harvesting, climate and geographical position, drying conditions, plant material, nature and polarity of the solvent and the method and modality of extractions.

The qualitative and quantitative analysis results show the superior biochemical quality of *Haloxylon scoparium*.

3.3 LC-MS-MS ANALYSIS RESULTS

The analysis results of *Haloxylon scoparium* leaf extract by LC-MS-MS are shown in Table 3. This analysis revealed the presence of several secondary metabolites. Twenty-three phenolic compounds were detected based on the LC-MS-MS in which fifteen were identified by comparison with standards. The results of this characterization confirm the presence of vanillin, naringenin, folic acid, maleic acid, benzoic acid, myricetin, quercetin, beta-carotene, butylhydroxy anisole (BHA), butylated hydroxytoluene (BHT), rutin, cafeic acid, hydroxy-4-coumarine, ascorbic acid, and gallic acid. However, keampferol, coumaric acid, picric acid, cinnamic acid, chlorogénic acid, chrysin, esculin, hesperetin were absent.

The phytochemical composition of *Haloxylon sco-*

Table 3: LC-MS-MS-determined phenolic compounds of *Haloxylon scoparium* Pomel. leaf extract

N°	Compound Name	Charge + / -	Precursor m/z	Product m/z	Haloxylon scoparium
01	Keampferol	$[MH]^+$	287.1	255.25	-
02	Vanillin	$[MH]^+$	153.10	71.15	+
03	Naringenin	$[MH]^+$	273.10	147.15	+
04	Coumaric Acid	$[MH]^+$	165.10	59.10	-
05	Picric Acid	$[MH]^-$	227.8	198.05	-
06	Cinnamic Acid	$[MH]^+$	149.1	77.2	-
07	Folic Acid	$[MH]^+$	442.90	323.45	+
08	Maleic Acid	$[MH]^+$	117.10	85.20	+
09	Benzoïc Acid	$[MH]^+$	123.10	91.20	+
10	Chlorogénic Acid	$[MH]^+$	355	73.15	-
11	Myricetin	$[MH]^+$	336.25	72.15	+
12	Quercetin	$[MH]^+$	303.10	85.05	+
13	Chrysin	$[MH]^+$	255.10	223.30	-
14	Esculin	$[MH]^+$	341.30	309.40	-
15	Hesperetin	$[MH]^-$	300.9	255.25	-
16	Beta-carotene	$[MH]^+$	537.20	199.25	+
17	Butylhydroxyanisole (BHA)	$[MH]^+$	181.10	140.15	+
18	Butylated hydroxytoluene (BHT)	$[MH]^+$	221	161.30	+
19	Rutin	$[MH]^+$	611.20	73.20	+
20	Cafeic Acid	$[MH]^-$	178.80	135.10	+
21	Hydroxy-4-Coumarine	$[MH]^-$	160.80	117.10	+
22	Ascorbic Acid	$[MH]^-$	174.90	131.10	+
23	Gallic Acid	$[MH]^-$	168.80	125.10	+

+ : present, - : not present

parium plant has not been the subject of many publications. Few researchers have investigated its phenolic composition. Chemical characterization of *Hammada scoparia* essential oils confirmed the presence of carvacrol, p-cymene, γ-terpinene and z-caryophyllene (Drioiche et al., 2019). In addition, Chao et al. (2013) showed the presence of some phenol acids such as Coumaric acid, Cinnamic acid and Caffeoylquinic acid, simple phenols (catechol and a chrysoeriol). However, Benkrief et al. (1990) identified isosalsoline dehydrosalsolidine, isosalsolidine (tetrahydroisoquinolines), N-methylcorydaldine (isoquinolone), tryptamine and N-methyltryptamine (tryptamines) as minor alkaloids. Other studies have isolated and identified two principal alkaloids: carnegine and N-methylisosalsoline from *Hammada scoparia* leaf extract (Jarraya et al., 2008; Bouaziz et al., 2016). A new flavonol triglycoside has been isolated from the leaves of *Hammada scoparia* (Salah et al., 2002).

3.4 PERCENTAGE OF GERMINATION INHIBITION

The allelopathic effect is expressed as the percentage of inhibition. The germination of target species, treated with *Haloxylon scoparium* leaf extract, decreased compared to the control. The degree of inhibition varies depending on the concentrations (Fig 1, 2, 3, and 4). A high inhibitory effect on germination was observed on all tested seeds, and the inhibition percentage increased with increasing concentrations of leaf extract. As illustrated in the graphs, at the 1 % and 2.5 % extract concentrations, the inhibition percentage values were, respectively, of 71.66-76.66 % for *Bromus rubens* , 63.33-93.33 % for *Phalaris minor* , 50.0-66.66 % for *Plantago lagopus* , 30.0-43.33 % for *Ammi visnaga* The 5 % and 10 % extract concentrations significantly inhibited the germination of weed seeds, which corresponds to a 100% inhibition percentage.

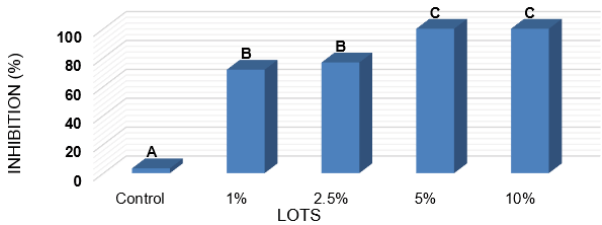

Figure 1: Inhibition percentage observed in control and treated lots by various concentration of leaf extract of *Haloxylon scoparium* Pomel. on *Bromus rubens* L. For each concentration, means (mean ± SD) followed by different letter (A, B, C) are significantly different at $p < 0.05$ level according to Tukey's LSD test

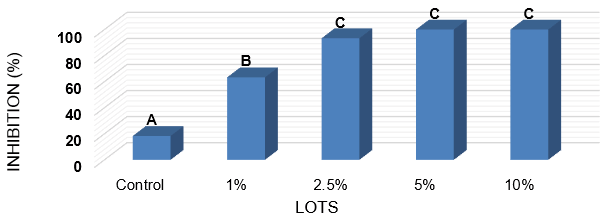

Figure 2: Inhibition percentage observed in control and treated lots by various concentration of leaf extract of *Haloxylon scoparium* Pomel. on *Phalaris minor* L. . For each concentration, means (mean ± SD) followed by different letter (A, B, C) are significantly different at $p < 0.05$ level according to Tukey's LSD test

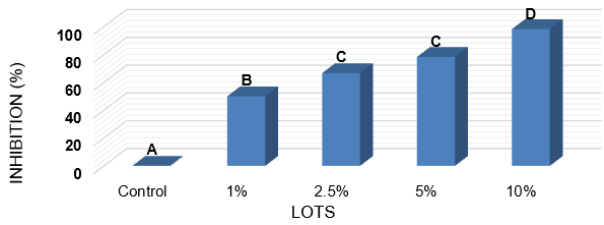

Figure 3: Inhibition percentage observed in control and treated lots by various concentration of leaf extract of *Haloxylon scoparium* Pomel. on *Plantago lagopus* L. For each concentration, means (mean ± SD) followed by different letter (A, B, C, D) are significantly different at $p < 0.05$ level according to Tukey's LSD test

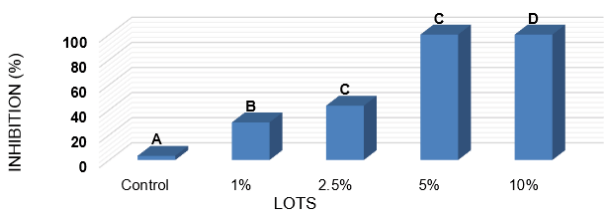

Figure 4: Inhibition percentage observed in control and treated lots by various concentration of leaf extract of *Haloxylon scoparium* Pomel. on *Ammi visnaga* L. For each concentration, means (mean ± SD) followed by different letter (A, B, C, D) are significantly different at $p < 0.05$ level according to Tukey's LSD test

According to the present research, *Haloxylon scoparium* leaf extract presents an allelopathic effect on the seed germination of weed species (*Bromus rubens* , *Phalaris minor* , *Plantago lagopus* , *Ammi visnaga*). These results are in agreement with those obtained by Karous et al. (2020); they demonstrate that the tested aqueous extract possessed an effective inhibitory activity against two weed species. Numerous studies have suggested that the presence of allelochemicals may cause a total or partial suppression of germination and a reduction in seedling growth (Qasem, 2002; Naz and Bano, 2013; Saadaoui et al., 2015; Mseddi et al., 2018). The presence of these secondary metabolites suggests that the plant might be of allelopathic and bioherbicidal importance.

3.5 AVERAGE GERMINATION TIME

In the present study, it was observed that the average germination time of weed species treated with leaf extract of *Haloxylon scoparium* increased in all treatments, from the lowest to the highest concentrations. The values show slower times compared to those of the control lot (Table 4). These results showed that the average germination time varies between 6.14 and 8.75 days.

The results of the current study show that *Haloxylon scoparium* leaf extract had an impact on average germination time. Da Silva et al. (2016) reported that *Ricinus communis* leaf extract significantly affected the average germination time, which increased with concentrations. Allelochemicals can increase cell membrane permeability, which prevents plants from absorbing nutrients from their environment and affects their normal growth (Li et al., 2010).

3.6 EFFECT OF EXTRACT ON SEEDLING GROWTH

The effect of *Haloxylon scoparium* leaf extract on the shoot and root growth of treated weed species (*Bromus rubens* , *Phalaris minor* , *Plantago lagopus* , *Ammi visnaga*) are shown in tables 5, 6, 7 and 8 respectively. In laboratory bioassay, all concentrations of leaf extract of *Haloxylon scoparium* decreased the seedling growth of weed species. They significantly decreased the shoot

Table 4: Effect of *Haloxylon scoparium* Pomel. leaf extract on average germination time of *Bromus rubens* L., *Phalaris minor* L., *Plantago lagopus* L. and *Ammi visnaga* L

Average Germination Time (AGT) (Days)								
Extract conc. (%)	Bromus rubens		Phalaris minor		Plantago lagopus		Ammi visnaga	
	Mean ± SD	Group	Mean ± SD	Group	Mean ± SD	Group	Mean ± SD	Group
Control	6.33 ± 0.11	A	7.31 ± 0.08	A	6.19 ± 0.05	A	6.53 ± 0.03	A
1 %	6.14 ± 0.16	A	7.91 ± 0.16	A	6.68 ± 0.36	AB	7.33 ± 0.05	B
2.5 %	6.54 ± 0.66	A	8.75 ± 1.44	A	7.13 ± 0.25	B	7.64 ± 0.29	C
5 %	-	-	-	-	7.85 ± 0.76	C	-	-
10 %	-	-	-	-	8,63 ± 0.47	D	-	-
LSD	0.72		1.51		0.72		0.30	

-:100 % inhibition percentage

Table 5: Effect of *Haloxylon scoparium* Pomel. leaf extract on shoot and root lengths and fresh mass of *Bromus rubens* L

Bromus rubens								
Extract conc. (%)	Shoot length (cm)		Root length (cm)		Shoot mass (g)		Root mass (g)	
	Mean ± SD	Group	Mean ± SD	Group	Mean ± SD	Group	Mean ± SD	Group
Control	12.61 ± 2.27	C	14.33 ± 1.49	C	0.0336 ± 0.0036	B	0.0262 ± 0.0078	B
1 %	5.45 ± 1.49	B	6.57 ± 1.05	B	0.0257 ± 0.0049	A	0.0096 ± 0.0024	A
2.5 %	2.12 ± 1.35	A	2.01 ± 1.05	A	0.0177 ± 0.0059	A	0.0058 ± 0.0020	A
LSD	3.16		2.47		0.0085		0.0043	

Table 6: Effect of *Haloxylon scoparium* Pomel. leaf extract on shoot and root lengths and fresh mass of *Phalaris minor* L

Phalaris minor								
Extract conc. (%)	Shoot length (cm)		Root length (cm)		Shoot mass (g)		Root mass (g)	
	Mean ± SD	Group	Mean ± SD	Group	Mean ± SD	Group	Mean ± SD	Group
Control	9.08 ± 0.65	B	9.26 ± 0.92	C	0.0155 ± 0.0019	B	0.0051 ± 0.0007	C
1 %	6.69 ± 0.87	A	3.97 ± 0.98	B	0.0096 ± 0.0018	AB	0.0018 ± 0.0009	B
2.5 %	5.45 ± 1.51	A	2.05 ± 1.41	A	0.0082 ± 0.0052	A	0.0005 ± 0.0001	A
LSD	1.93		1.92		0.0061		0.0011	

Table 7: Effect of *Haloxylon scoparium* Pomel. leaf extract on shoot and root lengths and fresh mass of *Plantago lagopus* L

Plantago lagopus								
Extract conc. (%)	Shoot length (cm)		Root length (cm)		Shoot mass (g)		Root mass (g)	
	Mean ± SD	Group	Mean ± SD	Group	Mean ± SD	Group	Mean ± SD	Group
Control	3.38 ± 0.49	D	4.03 ± 0.29	C	0.0084 ± 0.0015	C	0.0038 ± 0.0005	C
1 %	2.85 ± 0.29	C	1.53 ± 0.46	B	0.0058 ± 0.0024	B	0.0014 ± 0.0010	B
2.5 %	1.38 ± 0.25	B	0.28 ± 0.16	A	0.0026 ± 0.0021	A	0.0005 ± 0.0004	A
5 %	± 0.06	A	0.08 ± 0.03	A	0.0007 ± 0.0004	A	0.0001 ± 0.00005	A
10 %	0,35 ± 0.05	A	0,12 ± 0.05	A	0,0003 ± 0.0001	A	0,0001 ± 0.00005	A
LSD	0.38		0.41		0.0026		0.0009	

Table 8: Effect of *Haloxylon scoparium* Pomel. leaf extract on shoot and root lengths and fresh mass of *Ammi visnaga* L

Ammi visnaga								
Extract conc. (%)	Shoot length (cm)		Root length (cm)		Shoot mass (g)		Root mass (g)	
	Mean ± SD	Group	Mean ± SD	Group	Mean ± SD	Group	Mean ± SD	Group
Control	8.25 ± 1.03	B	6.49 ± 0.63	C	0.0251 ± 0.0047	A	0.0073 ± 0.0016	A
1 %	3.91 ± 0.79	A	1.73 ± 0.26	B	0.0083 ± 0.0012	A	0.0013 ± 0.0004	B
2.5 %	2.94 ± 0.45	A	0.59 ± 0.30	A	0.0063 ± 0.0026	A	0.0006 ± 0.0004	C
LSD	1.44		0.78		0.005		0.001	

length, root length, shoot fresh mass and root fresh mass of the test species compared to those of the control. This inhibitory effect on the root and shoot growth increased with increase of the concentrations.

The present study indicate that *Haloxylon scoparium* leaf extract presents allelopathic effect and contains allelochemicals responsible for the inhibitory activities on the germination and the seedling growth of test weed species. Other authors in their studies on weeds and crops observed similar findings (Scavo et al., 2018; Bhowmik and Doll, 1984). Allelochemicals affect plant germination and growth (Salhi et al., 2013). The capacity to inhibit seed germination and seedling growth are a complex process, and several hypotheses about allelochemicals of plant extracts have been formulated. These hypotheses suggest that these compounds might affect enzymes responsible for plant hormone synthesis, to inhibit the action of the amylase or inhibition of their tissue actions (Feeny, 1976). The alteration of the synthesis or activities of gibberellic acid in the seed could be due to the presence of phenolic compounds (Olofsdotter, 2001). Cell division and elongation are susceptible to the presence of allelopathic compounds (Muller 1965), resulting in the reduction of root and shoot growth (Qasem, 2002).

4 CONCLUSION

The results of the present research confirmed the strong allelopathic effects of *Haloxylon scoparium* leaf extract, on the germination, shoot and root growth of tested weed species (*Bromus rubens*, *Phalaris minor*, *Plantago lagopus*, *Ammi visnaga*). The phenolic compounds present a great interest for the researchers due to their various biological activities. These findings encourages future research for identifying and characterizing germination and growth inhibitors; it could be the source of these species' significant allelopathic potential. These allelochemicals maight be used in the research and development of weed-controlling environmental herbicides**.**

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