

## **Influence of extracts of *Chenopodium album* and NaCl salinity on germination and seedling growth of soybean**

A. NAMVAR<sup>\*1</sup>, R. SEYED SHARIFI<sup>2</sup>, T. KHANDAN<sup>1</sup> and P. MOLAEI<sup>1</sup>

1. Members of Young Researcher Group of Iran Islamic Azad University, Ardabil Branch

2. Faculty of Agronomy and Plant Breeding, College of Agriculture, University of Mohaghegh Ardabili, Ardabil, Iran.

E. Mail: namvar\_a60@yahoo.com

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### **ABSTRACT**

Under lab conditions, bioassay studies were done to study the allelopathic effects of aqueous extracts (0,10,20,40%) of different parts (leaf, root, leaf + root) of common lambsquarters (*Chenopodium album* L.) and salinity (0, 75 and 150 mM NaCl) on germination of soybean (*Glycine max* L.). The different types and concentrations of *C. album*. extract and salinity adversely inhibited all germination traits of soybean. The aqueous leaf extracts of *C. album* showed the highest inhibitory effects on studied germination traits of soybean, followed by root + leaf and root extracts, respectively. The degree of inhibition increased with increasing concentrations of extract and salinity. The extracts + NaCl drastically reduced the growth more than the separate effects of these stress treatments e.g., extract and salinity alone. The highest concentrations of extract and salinity reduced the radicle length by 63 and 72% respectively, while leaf extract + NaCl at highest concentrations caused 88% reduction than control. Radicle weight and plumule length and weight showed similar response.

**Key words:** Allelopathy, *Chenopodium album*, Common lambsquarters, germination traits, salinity, soybean.

### **INTRODUCTION**

Interference occurs when one plant species fails to germinate, grows more slowly and shows damage or does not survive in the presence of another plant species. Such interference can result from competition and allelopathy. Allelopathy is the negative effect of chemicals released by one plant species on the growth or reproduction of another (20). Allelopathy plays major role in both natural and agro-ecosystems. It may be used to improve the crop productivity and environmental protection (through eco-friendly control of weeds, pests, crop diseases) and nitrogen conservation in crop lands (10). Allelochemicals are present in all plant tissues (leaves, flowers, fruits, stems, roots, rhizomes, seeds and pollen) and are released through volatiles, leachates, exudates from living roots or decay of biomass (19,22). Information is available about the allelopathic effects of weeds on crop plants, crop plants on weeds, weeds on weeds or crops on crops (15). However, the information on the allelopathic influence of weeds in some crops is lacking.

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\*Correspondence Author.

The *Chenopodium album* L. is most obnoxious weed in many countries and is common in wheat, barley, sugarcane, soybean and vegetable fields (2). The aqueous extracts of its leaves or residues inhibits the germination and growth of other plant. Its root exudates retarded the growth of shoot and root of corn and soybean in a nutrient solution (7). Its aqueous extracts significantly reduced seed germination, shoot and root length in wheat (6), radicle growth in corn, bean and squash (3), hypocotyls growth in soybean, wheat and radish (16), reduces shoot and root length and weight in wheat (2) and growth of cucumber plant (23). Salinity is major problem world wide (9) similar to adverse effects of *C. album* on growth of crop plants and also adversely effects the growth of plants. It prevents the seed germination (11,14) and seedling growth (9,13). The response of legumes to salinity varies greatly and depends on climatic conditions, soil properties and stage of plant growth. Some legumes (*Vicia faba*, *Phaseolus vulgaris* and *Glycine max*) are more salt tolerant (8). Mensah *et al.* (17) reported that germination, seedling growth and radicle elongation of groundnut decreased with increasing salinity. The dry weight of wheat genotypes shoot and root decreased with increasing salt concentration (5), likewise, salinity significantly decreases the radicle and coleoptile length in barley (9).

The presence of weed and salinity together may be more hazardous to crops growth than weed and salinity alone. Alam *et al.* (2) showed that the combination of leaf extract of common lambsquarters and NaCl drastically reduced the growth of shoot and root more than the separate effects of these stress treatments in wheat. However, most of the studies were done under normal growth conditions and there are no reports on allelopathic effects of weeds on crop plants, growing under saline conditions. Therefore, this study aimed to demonstrates the effects of aqueous extracts of different parts of common lambsquarters (*Chenopodium album* L.) in the presence of different concentrations of salinity on germination traits of soybean (*Glycine max* L.).

## MATERIALS AND METHODS

The lab bioassay studies were conducted in Seed Technology Laboratory during 2008. The treatments were replicated thrice in randomized complete block. There were 3 Experimental factors: (i). Aqueous extracts: 3 (leaf, root and leaf + root), (ii). Extracts concentrations: 4 (0, 10, 20 and 40%) and (iii). Salinity levels: 3 (0, 75 and 150 mM NaCl). The fresh leaves and roots of *C. album* were randomly collected at vegetative stage, then washed thoroughly with distilled water and dried in oven at 70°C for 24 h. The dried leaf and root samples were grounded in a Willey mill, separately to pass through a 20 mesh screen. The aqueous leaf and root extracts were prepared by soaking 10 g powdered leaf and root materials in 100 ml distilled water for 24 h. Root + leaf extract were obtained by soaking 5 g root and 5 g of leaf in 100 ml water. Thereafter, these were filtered using No.1 Whatman filter paper. From these leachates, 3 concentrations (10, 20 and 40%) of leaf, root and root + leaf extracts were prepared by diluting with distilled water. Mixture of extract and NaCl solution was prepared by adding equal ratio of each solution. Distilled water was control in all treatments.

The soybean cv 'Williams' seeds were surface sterilized with 1% sodium hypochloride for 10 min, then rinsed with distilled water several times. 15 seeds of soybean were placed in sterilized petri dishes lined with two layers of Whatman No.1 filter

paper. These petri dishes were saturated with respective leachate using 5.0 ml solution. Moisture in the petri dishes was maintained by adding 1.0 ml extracts, mixture of extracts and NaCl solutions or distilled water as required. Petri dishes were placed in a growth-chamber (day/night temperature of 24°/15°C and 16 h photoperiod).

Germination of all seeds was recorded till 3-days, the number of germinated seeds were consistent. Equations 1 and 2 and their parameters were used to express the germination situation.

$$G = S_{NG} / S_{NO} \times 100\% \quad (1)$$

Where, G is germination capacity (%),  $S_{NG}$  is the number of germinated seeds and  $S_{NO}$  is the number of experimental seeds with viability (12).

$$GR = S_{Nm} / S_{NO} \times 100\% \quad (2)$$

Where, GR is the germination rate (seeds day<sup>-1</sup>),  $S_{Nm}$  is the number of germinated seeds at the germination peak (generally one third of the way through the germination period) (24).

Seedling vigour index (SVI) was calculated as per following modified formula of Abdul-Baki and Anderson (1).

$$SVI = \text{Germination capacity (\%)} \times \text{Radicle length (cm)} \quad (3)$$

Besides G, GR and SVI, the measurements were done of length and dry weight of radicle and plumule. Then, ratio of radicle weight to plumule weight was calculated.

The allelopathic activity was expressed as inhibition rate, calculated by using following equation:

$$IR = (L_c - L_t) / L_c \times 100 (\%) \quad (4)$$

Where, IR is the inhibition rate (%),  $L_c$  is the radicle or plumule length (or weight) of a control plot and  $L_t$  is the radicle or plumule length (or weight) of a test plot.

**Statistical Analysis:** Analysis of variance was done using SAS computer software package and the mean values were compared with least significant difference (LSD) test. Variances were homogeneous (testing with Bartlett's test) and non additive (testing with Tukey's test).

## RESULTS AND DISCUSSION

### Germination, germination rate (GR) and seedling vigor index (SVI)

The extract type (leaf, root and root + leaf) had no significant effect on germination and GR. However, increasing extract and salinity concentrations significantly decreased the germination and germination rate (Table 1). The highest concentration of extract (40%) decreased the germination by 15% and GR by 43%. Similarly, the maximum salinity level (150 mM) decreased the germination and GR by 23 and 53% respectively,

compared to control. The leaf extract showed the lowest SVI than root and root + leaf extracts. The increasing extract and salinity concentrations decreased the germination GR and SVI. The highest extract concentration (40%) and salinity (150 mM) decreased the SVI by 69 and 78% respectively, compared to control (Table 1).

Table 1. Effects of aqueous extracts of *Chenopodium album* L. extract concentrations and salinity on germination and seedling growth of soybean

| Treatments                        | Germination (%) | Germination rate (seeds day <sup>-1</sup> ) | Radicle length (cm) | Plumule length (cm) | Radicle weight (mg) | Plumule weight (mg) |
|-----------------------------------|-----------------|---|---------------------|---------------------|---------------------|---------------------|
| <b>Extract type</b>               |                 |   |                     |                     |                     |                     |
| Root <sub>ex</sub>                | 83.75 a         | 0.47 a                                      | 3.1 a               | 3.2 a               | 7.0 a               | 6.6 a               |
| Leaf <sub>ex</sub>                | 83.00 a         | 0.46 a                                      | 2.7 c               | 2.8 c               | 5.6 b               | 5.3 c               |
| Root/Leaf <sub>ex</sub>           | 83.67 a         | 0.47 a                                      | 2.8 b               | 3.0 b               | 6.8 a               | 6.1 b               |
| <b>Extract concentrations (%)</b> |                 |   |                     |                     |                     |                     |
| 0                                 | 90.11 a         | 0.60 a                                      | 4.0 a               | 4.3 a               | 9.6 a               | 9.6 a               |
| 10                                | 86.00 b         | 0.52 b                                      | 3.5 b               | 3.7 b               | 7.9 b               | 6.9 b               |
| 20                                | 82.00 c         | 0.45 c                                      | 2.6 c               | 2.7 c               | 5.0 c               | 4.7 c               |
| 40                                | 75.78 d         | 0.34 d                                      | 1.5 d               | 1.3 d               | 3.3 d               | 2.9 d               |
| <b>Salinity levels (mM)</b>       |                 |   |                     |                     |                     |                     |
| 0                                 | 94.08 a         | 0.63 a                                      | 4.3 a               | 4.5 a               | 9.1 a               | 9.6 a               |
| 75                                | 84.33 b         | 0.51 b                                      | 3.2 b               | 3.2 b               | 6.3 b               | 5.5 b               |
| 150                               | 72.00 c         | 0.29 c                                      | 1.2 c               | 1.2 c               | 4.0 c               | 2.9 c               |
| Mean                              | 83.47           | 0.47  | 2.894               | 2.995               | 6.485               | 6.029               |

Means followed by same letters in each treatment showed no significant difference at 5% level by LSD test.

The decrease in seed germination or seedling development causes significant reduction in yield (20). The allelochemicals (6,20,22) and salinity (9,17) are harmful to germination. The inhibitory effects of salinity on seed germination may be due to low water potential, ion toxicity and ion imbalance (17). The released toxic metabolites from different plant parts also adversely affects the germination (18). Such metabolites inhibited and/or delayed the seed germination and stunted the growth. Delay seed germination owing to allelopathic extract or salt solution could be confounded with osmotic effects on rate of imbibition, delayed initiation of germination and especially cell elongation, the factor that affects root growth before and after the tip penetrates the seed coat (10).

SVI reflects seed the vigour under different stress conditions (1). It was decreased due to reduction in radicle length and germination, affected by salinity and allelopathic effects of lambsquarters extracts (Table 1).

### Seedling growth

All treatments (extract type, extract concentration and salinity) significantly reduced both the radicle and plumule lengths of soybean (Table 1). The leaf extracts were most inhibitory to radicle and plumule length followed by root + leaf and root extracts. The highest concentration of extract (40%) decreased the radicle and plumule length by 63 and 71% respectively, than control (Table 1). Likewise, 75 and 150 mM NaCl reduced the radicle length by 26 and 72% and plumule length by 28 and 74% respectively, over the

control (Table 1). The increasing concentrations of all extracts (leaf, root and root + leaf) significantly decreased the radicle and plumule length, but increasing concentrations of leaf extract were most inhibitory (Figure 1). Also, the minimum radicle and plumule lengths in each levels of salinity recorded at the maximum concentrations (40%) of lambsquarters extracts (Figure 2). Furthermore, leaf extract caused more reduction in radicle and plumule length with increasing salinity concentration. The leaf extract with 150 mM NaCl reduced radicle and plumule length by 88 and 90% respectively, compared to control (Figure 3).

The aqueous leaf extract was inhibitorier to radicle and plumule weight than the root and root + leaf extracts. Leaf extract reduced the radicle and plumule weight by 19 and 16%, than root and root + leaf extracts, respectively (Table 1). The degree of inhibition increased with the increasing concentrations of extract and salinity. The maximum concentration of extract (40%) reduced the radicle and plumule weight by 65 and 69% over the control. Likewise, the NaCl levels (75 and 150 mM) reduced radicle weight by 30 and 55% and plumule weight by 42 and 69% than control (Table 1). The increase in concentration of leaf extract reduced the radicle and plumule weight more than root and root/leaf extracts (Figure 1). The highest concentration of extracts caused maximum reduction in radicle and plumule weight at the highest concentration of salinity (Figure 2). Furthermore, the leaf extract adversely affected the radicle and plumule weight with increasing salinity concentrations. The leaf extract with 150 mM NaCl reduced the radicle and plumule weight by 77 and 82% respectively, over control (Figure 3).

In present study, lambsquarters extracts (leaf, root and root + leaf) had allelopathic effects on radicle length and weight and plumule length and weight, but leaf extract was most inhibitory to germination traits (Table 1). Because leaves are more allelopathic than other plant tissues (3,16,20). More inhibitory effects of leaf extract may be due to the fact that allelochemicals mostly synthesize in the roots and are stored in vegetative parts such as leaves (3). The increasing concentrations of extract and salinity significantly decreased the radicle length and weight and plumule length and weight (Table 1). Our results are in agreement with Bhowmik and Doll (7) in corn and soybean, Anaya *et al.* (3) in corn, bean and squash, Mallik *et al.* (16) in soybean, wheat and radish, Alam *et al.* (2) in wheat and Tamaki *et al.* (23) in cucumber.

Numerous bioassay studies had proved that different plant parts release toxic metabolites that adversely affect the germination and growth of crops. Besides, allelochemicals may indirectly inhibit the water uptake (18).

Salinity generally negatively affects the growth and development of plants. The negative effects of salinity on germination and growth occurs especially in osmotic process, hence, seeds cannot absorb enough water to germinate. Furthermore, salinity strongly affects the cell membrane and its stability. Borucki and Sujkowska (8) reported the inhibitory effects of salinity on growth of pea treated with 75 mM NaCl. NaCl reduces the radicle and plumule length in barley (9). Bagci *et al.* (5) studied the effects of salinity on different genotypes of wheat and observed reduction in root and shoot dry weight with increasing salinity concentration in all genotypes. Our findings also have similar results (Table 1).

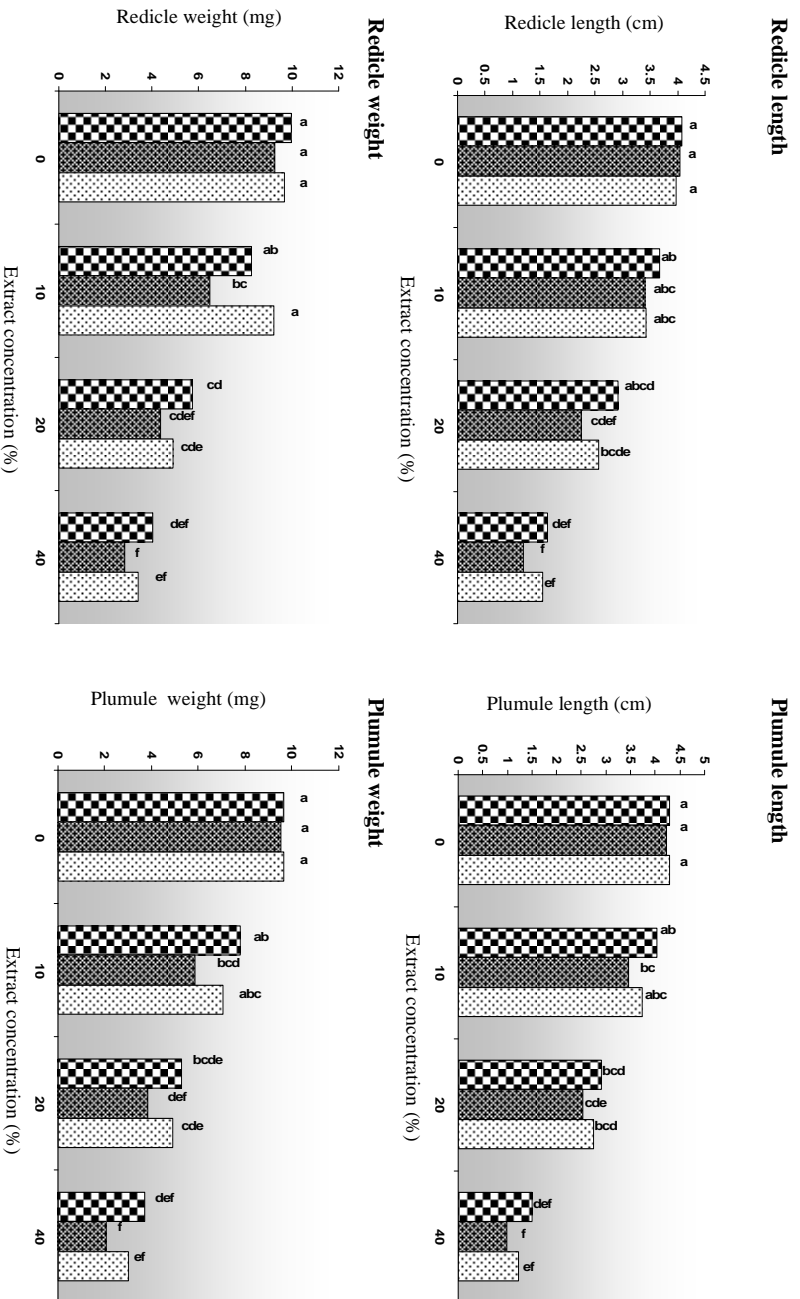


Figure 1. Effect of different extracts concentrations (0, 10, 20 and 40%) and extracts types (root, leaf and root/leaf) of lambquarters on radicle length, plumule length, radicle weight and plumule weight of soybean. Bars with the same letters are not significantly different (LSD test at 5% level).

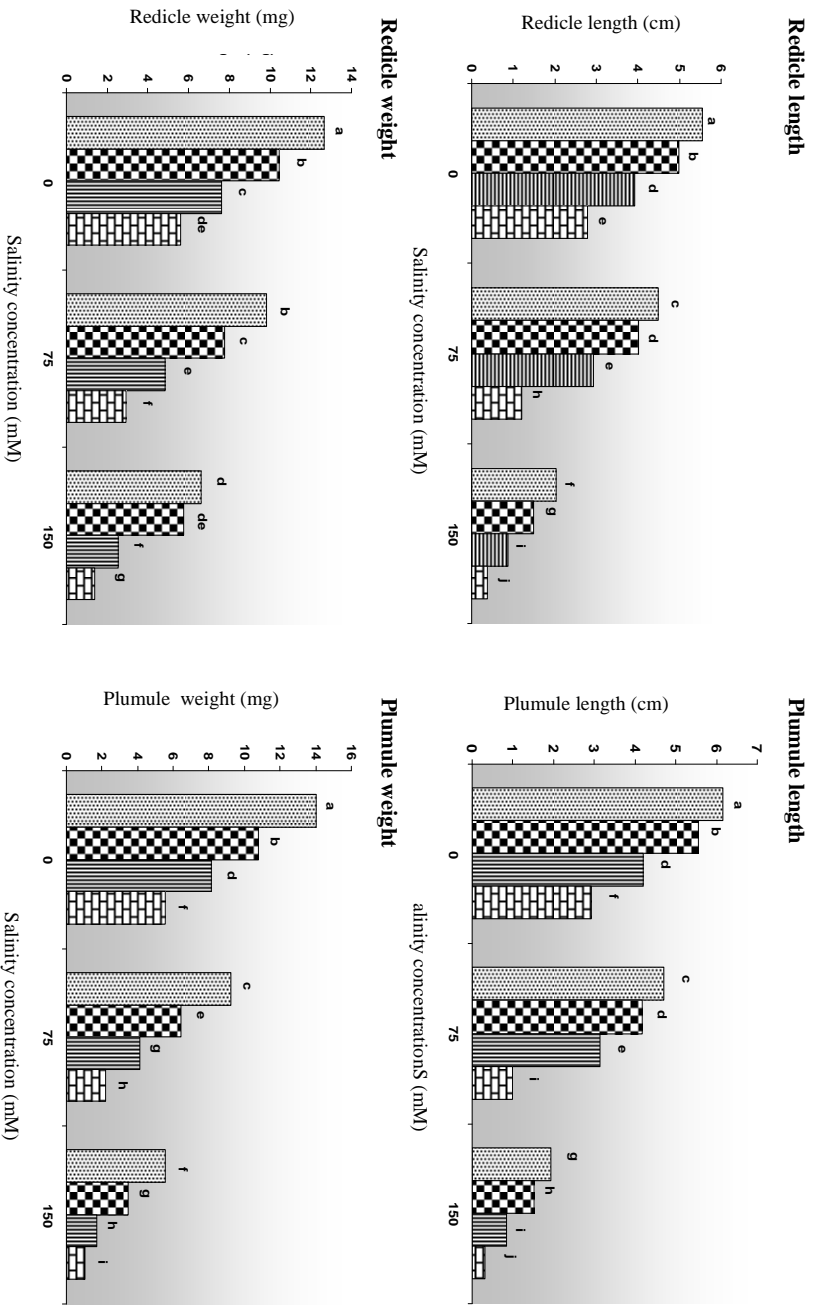


Figure 2. Effect of salinity levels (0, 75 and 150 mM NaCl) and lambdaguars extract concentrations (0, 10, 20 and 40%) on radicle length, plumule length, radicle weight and plumule weight of soybean. Bars with the same letters are not significantly different (LSD test at 5% level).

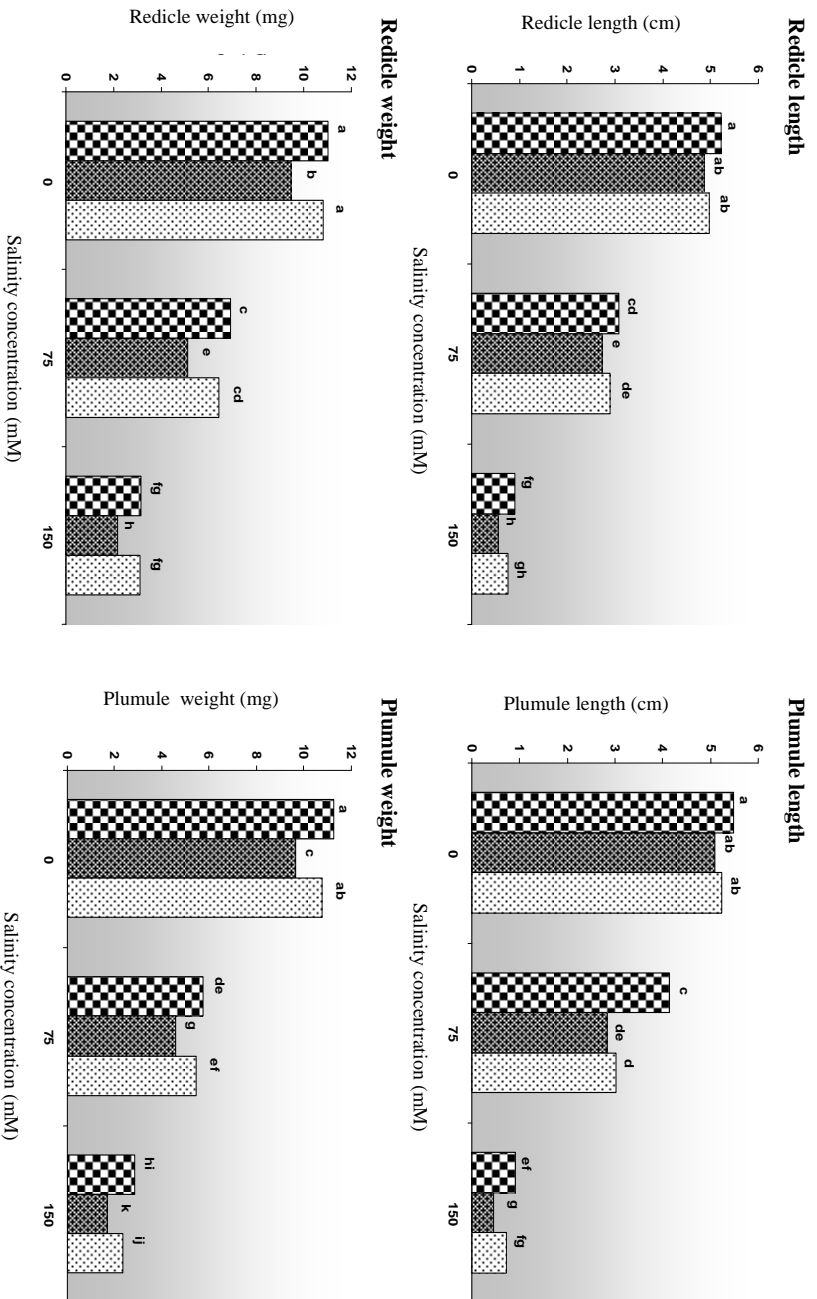





Figure 3. Effect of different salinity levels (0, 75 and 150mM NaCl) and extract types (root , leaf , and root/leaf ) of lambsquarters on radicle length, plumule length, radicle weight and plumule weight of soybean. Bars with the same letters are not significantly different (LSD test at 5% level).

Allelopathic phenomenon is also strongly coupled with other stresses of crop environment (temperature extremes, nutrient and moisture variables). These stresses often enhance the allelochemicals interference. The crops are more sensitive to allelopathy under unfavourable moisture, temperature or nutrient conditions. The salinity increases the effects of allelochemicals and both exert deleterious effects (2). The combination of extract and NaCl, drastically reduced the growth more than their separate effects (Figure 3) e.g., extract and salinity alone at the highest concentrations reduced the radicle length by 63 and 72% respectively (Table1), while the leaf extract + NaCl at the highest concentration decreased radicle length by 88%, compared to control (Figure 3). There is little evidence on the effects of salinity in combination with common lambsquarters, because *C. album* is not major weed in saline soils. The salinity + lambsquarters extract reduced the radicle length and weight and plumule length and weight more than the extract and/or salinity alone (2).

The plumule length and weight were more affected than root (Tables 1), because the turgor pressure is different in each part of plant. For instance, the turgor pressure for growth of wheat root and leaf are 0.05 and 0.1 MPa, respectively. Therefore under same water stress, the growth of one part of plant may be inhibited, while, other parts grew (21). Furthermore, increase in radicle weight: plumule weight ratio with increasing extract and salinity concentration (Table 1) showed more decrease in plumule weight than radicle weight.

## CONCLUSIONS

Lambsquarters aqueous extracts (leaf, root and root + leaf) had allelopathic effects on germination traits, however, leaf extract proved more inhibitory than root and root + leaf extracts. Furthermore, the degree of inhibition increased with increasing concentrations of extract and salinity. The soybean radicle was more sensitive than plumule. Moreover, the extract + NaCl drastically reduced the growth more than separate effects of these stress treatments, thus more attention should be paid to allelopathic effects of weeds on crops in saline soils.

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