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THE EFFECT OF SALINITY AND NITROGEN ON GROWTH AND MORPHOLOGICAL CHARACTERISTICS OF DILL

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ABSTRACT

Dill (Anethumgraveolens L.) is an annual and aromatic plant belonging to the umbel variety (Apiaceous) which is used in food and pharmaceutical industries. In order to investigate the interactional effect of salt (NaCl) and nitrogen (NH_4No_3) on the growth and physiological characteristics, an experiment was conducted in 2011 in the faculty of Agriculture, University of Lorestan. It was performed in a factorial design with completely random blocks in six replications and 15 treatments. Seeds of this plant were planted in the pot with a diameter of 25 cm soil containing a mixture of three parts, cultivar soil, sand and manure. In eight leaf - stage, salt treatment (NaCl) at concentrations of 0, 30, 60, 90, 120gr of sodium chloride in per 100 kilograms of soil and ammonium nitrate treatment with different concentrations of 0, 10, 20gr per 100 kg were applied. The results of the variance analysis suggested that increased levels of salinity and ammonium nitrate has a significant effect on the parameters measured (P≤0/01). With increasing salinity levels, relative growth rate, fresh weight and dry, main stem length, fresh and dry weight of shoot and root decreased while applying ammonium nitrate in the shoot and root length, fresh and dry weight of root and shoot, the RGR , dry weight in the root and aerial organs were decreased.

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KEY WORDS

Dill, sodium chloride, ammonium nitrate.

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INTRODUCTION

Salinity stress is one of the most common abiotic stresses that decrease agricultural production in arid and semi-arid lands and use efficiency. According to the Food and Agricultural Organization (FAO), about 300 million hectares of farmland worldwide is under salt stress [1]. Salinity is the accumulation of soluble salts in the soil depth or around the roots of plants, to the extent that it affected the growth and development and leads to death. Although the salt stress can refer to any salt, but usually the purpose of salinity, is concentration of sodium chloride which is the most abundant salt in the nature. According to botanists, salinity stress causes many problems such as water scarcity and physiological drought, morphological changes such as reduced growth, reduced leaf area, fleshy leaves, thick cuticle, changes in the number and size of pores, creating Tyloses, woody roots and shorten plant [2], poisoning of the sodium and chlorine elements [3], impaired absorption of other ions [4] and biochemical changes such as the production of reactive oxygen species [1]. On the one hand, according to many properties of medicinal plants and humans need to use these drugs, effective production of them has always had a great importance. Dill is the among the herbs and nutritional plants that its production for pharmaceutical products, essential oils and health is of great importance. Considering the fact that in Iran, 12 percent of the cultivated land area is affected by salinity close to 50% [5], identification of medicinal plants tolerant to salinity and their increased production efficiency in these areas are essential. In addition, the effect of nitrogen on the growth and root development provides necessary conditions for water uptake and other nutrients and inhibits the excessive loss of photosynthesis process and stomatal conductance under salt stress conditions [6]. Therefore, in this article, with respect to the objectives and assumptions, we investigated the effect of salinity and nitrogen on the growth and physiological characteristics of dill in detail.

Research objectives

1. The effect of salinity on growth and development of physiological characteristics of dill plant.
2. The effect of nitrogen on the growth and development of physiological characteristics of dill plant.

3. The effect of nitrogen on the growth and physiological characteristics of dill under salt stress.

Salinity is effective in the biochemical and physiological of dill plant. Nitrogen is effective in the biochemical and physiological of dill plant. We hope that the results of this research opens the way for future research within the area of botany regarding the optimal production of plant products, particularly pharmaceutical products.

Effect of salinity on seed germination and plant growth parameters

Salinity is generally causes in delayed germination and rate of germination. Depending on the severity of it, salinity can cause lack of germination in some seeds, or delayed germination or weak production in the plants. In germination stage of seed, some plants such as wheat and barley show low tolerance to salinity yet their tolerance is increased at later growth stages [7]. The investigation of the effects of salinity on dill indicates that in the five ds salinity, germination was decreased to 50% and is stopped in 35 dS concentration, and also grain production in salinity five ds was reduced and inhibited at concentration of 28 dS [8]. Through the facilitation of absorption of toxic ions, which alter the activity of certain enzymes and hormones, salinity affects the germination of seeds. These physical and chemical changes result in reduced germination. Salt (sodium chloride) and polyethylene glycol (PEG) both reduce the absorption rate, but the latter is more pronounced [9]. By examining five salinity treatments (100, 75, 50, 25, 0 mM) in the pot, Nooraniazad and Hajibagheri (2008) found that increasing salinity treatments led to reduced leaf area and pot, total dry material and stem length in dill. Several parameters are effective in salt tolerance of plants, including survival rate, plant height, leaf area, leaf damage and their relative growth rate [10]. The most important reason for the study of soil salinity is because it's effect on plant growth and yield and its affiliated parameters. Sodium chloride salinity is the most common types of salinity in the arable soils of Iran which limits the capability to produce many important plants. Species and different varieties of plants show different resistances under the same saline conditions or at the certain stage of growth. The harmful concentration of salt to different plants depends on the salts mixture, soil characteristics, climate, humidity and plant [4]. Tolerance between different species of plants to salinity not only is different but also depends on the environmental conditions. The effect of salinity takes place on carbon accumulation capacity primarily by reducing the level of photosynthesis in the leaf [11]. Because of reduced osmotic potential effect in the root, the absorption of water and minerals is decreased by plant. Also due to a disorder in the process of photosynthesis, the plant growth, leaf, stems length and branches production are reduced [12]. In studies of the tomato plant in salt concentration of 70 and 90 mm, it was shown that, fresh and dry weight of the plant was decreased by increasing the salt concentration.

The effect of nitrogen on plants in saline conditions

Nitrogen is among the factors that are necessary to meet the requirements for the plant [13]. Significant effects of nitrogen in the increased crop yields as well as reduced the amount of soil nitrogen motivated researchers to use nitrogen fertilizers for increased efficiency of their culture. Culture and access to factors, in order to increase the medicinal properties of plants were always considered important for the people involved in the pharmaceutical industry, including the effects of chemical fertilizers such as ammonium nitrate in the quantity and quality of the materials. The treated seeds of an onion with ammonium nitrate or potassium nitrate under salinity stress, its resistance and germination were increased. Salinity, amount of tissue, also reduces the length and weight of roots. In addition to osmotic effect, root to shoot ratio is increased. The growth of greenhouse under salt stress depends on the concentration of nitrogen used. The seeds of many plants and shrubs show higher yields in the presence of nitrates to ammonia [14]. Plant treatment with ammonium nitrate under saline condition increases biomass and plant growth. The effect of nitrate on shoot dry weight and growth is more tangible. Dill has the ability to absorb nitrates from the dry soil, resulting in decreased harvest index and biomass production is increased. Nitrate increases dry weight of leaf and root dry weight while used alone or in conjunction with ammonium. Onion performance significantly increases with the use of nitrogen fertilizers (Urea), as well as leaf and dry weight of onion significantly increases with increasing N rate. Many environmental stresses such as salinity, have an adverse effect on the uptake and assimilation of nitrogen, some studies suggest that salinity reduces absorption and the accumulation of nitrogen in the aerial parts of the plant [15].

MATERIALS AND METHODS

First test

Germination

The first experiment was carried out at temperatures between 15 to 21°C. After preparing healthy and uniform seeds, they were put in 20% sodium hypochlorite solution for 10 minutes, and then were washed with water several times. Then two pieces of filter paper (Whatman) were placed in each Petri dish (diameter 70 mm), and 10 ml of sodium chloride solution at a concentration of 2, 4, 6, 8 and 10 per cent was poured into them.

In order to germinate, Petri prepared were located in a cool place with temperatures below 20 degrees Celsius based on the above method. The number of buds per Petri was recorded at intervals of a day the traits assessed included the following: Germination percentage and speed were calculated through the following formulas: Germination percentage = the total number of germinated seeds until the last day of the counting of 100÷the total number of seeds

Germination speed = germinated seeds ÷first day of counting + the second day of counting + the number of germinated seeds ÷last day of counting

The second test

Dill cultivation

During the winter in 2011 and spring in 2012, a pot experiment in a completely randomized design with 15 treatments, were performed in the greenhouse of Lorestan University (over 90 vases in five kilograms) which every treatment consisted of six pots. In each pot, equal amount of the soil (three-fifths), sand (one-fifth) and urea (one-fifth) was poured. Dill seeds were disinfected with 30% commercial sodium hypochlorite before planting for five minutes, and then were washed with distilled water during three successive stages. 30 seeds were planted in each pot to a depth of three centimeters which were sparse at regular distances in the different stages of plant growth.

The application of different treatments on plants

Seeding was cultivated in early February, approximately 45 days after the plant was reached in eight leaf stage. Salinity treatments and ammonium nitrate were applied. Then, factors such as shoot, root length, leaf number, fresh and dry weight were randomly measured from several pot plants. In Table (2-3) the application of different levels of sodium chloride in the pots was plotted. The numbers presented in Table were obtained by multiplying the treatments product (0, 30, 60, 90, and 120) in 5 divided by 100. The following **Table -3.1** shows that the application of stress is gradually conducted to avoid shock to the plant.

Table: 3.1: Weekly schedule for the application of salinity stress in grams per 100 kg of soil

Treatment	First week	Second week	Third week	Fourth week
Control	0	0	0	0
30g/100kg Soil	1.5	0	0	0
60g/100kg Soil	1.5	1.5	0	0
90g/100kg Soil	1.5	1.5	1.5	0
120g/100 kg Soil	1.5	1.5	1.5	1.5

In **Table- 3.2**, the application of different levels of ammonium nitrate has been plotted. The resulting numbers are derived by the formula.

Table 3.2:Weekly schedule for the application of ammonium nitrate in grams per 5 kg of soil

Treatment	First week	Second week	Third week	Fourth week
Control	0	0	0	0
10g/100 kg Soil	0.5	0	0	0
20g/100 kg Soil	0.5	0.5	0	0

Growth Analysis

In order to study plant growth, the number of 15 plants was randomly chosen from the pot 45 days after treatment. Then their roots and shoots were dried well after washing and their stem, roots, leaves and inflorescence umbrella were calculated in grams to two decimal points. As well, their length of the stem, branches and roots were calculated in terms of centimeters to one decimal point, and then dry weight. Other developmental parameters were calculated in the following way:

1) Relative growth rate (RGR): unit is gr.Kg-1.d-1.

$$\mathrm{RGR} = \frac{\ln W_2 - \ln W_1}{T_2 - T_1}$$

2) ln denotes a Nehpri logarithm. W_1 and W_2 are the total dry weights at the beginning and end of the experiment, respectively. T1 denotes the primary time (45 days) and T2 to the secondary time (75 days).
3) The ratio of root dry weight to shoot dry weight is called alometric coefficient which is considered as indicators of water plant.
4) R/S = shoot / root dry weight
5) It is worth mentioning that the statistical analysis of the impact of ammonium nitrate and sodium chloride were carried out separately and together. For this purpose, 9.01 SAS statistical software was used. Mean comparisons were calculated using Duncan's test results listed in the following section.

RESULTS

The first test results

Germination percentage changes in different treatments of NaCl

The results of this study showed that increasing concentrations of NaCl decreased the germination and reduced early seedling growth of plant so that these changes were significant at one percent level. In the salinity treatment, the highest germination percentage (68%) in the control group and the minimum germination (zero) in a 10% concentration of salt (sodium chloride) were observed, respectively.

Changes in germination rate in different treatments of NaCl

Increasing the concentration of NaCl in the experimental groups reduced the rate of germination in the plant, so these changes were significant at the %1 probability level. The highest germination rate (35/03%), and the lowest germination rate (zero percent) were observed in the control group and 10% NaCl, respectively. In ammonium nitrate treatment, the highest germination rate (35/03%) was observed in the control group with zero concentration and the lowest germination rate (zero percent) was observed in the treatment groups at a concentration of 10% ammonium nitrate.

The average length of root and shoot

After 10 days, the average length of root and shoot was measured. The maximum root length (4.3cm) in the control group (with zero-percent concentration of sodium chloride), and the minimum length of root (zero) in 10 percent concentration of sodium chloride were observed, respectively. The highest shoot length (2.7cm) in control group (with zero-percent concentration of sodium chloride) and the lowest shoot length (zero) in chloride concentration 10% were observed, respectively. The results showed that there is a significant difference between different treatments of salinity on the average length of root and shoot at the %1 probability level. In this study, speed changes and germination percent in different concentrations of ammonium nitrate were zero except control group, so the results were not mentioned.

The second test: Effect of salinity and nitrogen on the growth and morphological characteristics of dill

Root length changes

Average root length in the effect of sodium chloride and ammonium nitrate salt was significantly changed at %1 probability level, so that in the comparison test of Duncan means, the highest root length (35/57 cm) and the minimum length of root (2-10 cm) were observed, respectively.

Changes along the main stem

The average length of the main stem of the effect of salt and the ammonium nitrate showed no significant changes at %1 probability level so that in the comparison test of Duncan means, the maximum length of main stem (70/6cm) and the shortest main stem (12-1cm) were observed, respectively.

Changes in the shoot's fresh weight

The average shoot fresh weightin the different treatments of the ammonium nitrate and sodium chloride showed significant change at %1 probability level so that in the comparison test of Duncan means, the maximum fresh weight of stem(640g) and the lowest weight (253g) were observed, respectively.

Changes in the shoot's dry weight

The average changes in dry weight of aerial organs in the different treatments of the ammonium nitrate and sodium chloride showed significant change at %5 probability level so that in the comparison test of Duncan means, the maximum dry weight of stem(104g) and the lowest dry weight of the stem (41g) were observed, respectively.

Change in the root's wet weight

The average wet weight of root in the different treatments of the ammonium nitrate and sodium chloride showed significant change at %1 probability level so that in the comparison test of Duncan means, the maximum wet weight of root (708g) and the lowest dry weight of the root (328g) were observed, respectively.

Change in the root's dry weight

The average dry weight of root in the different treatments of the ammonium nitrate and sodium chloride showed significant change at %1 probability level so that in the comparison test of Duncan means, the maximum dry weight of root (14/9 g) and the lowest dry weight of the root (5/9 g) were observed, respectively.

Relative Growth Rate (RGR)

Relative growth rate with increasing concentration of sodium chloride and ammonium nitrate in the pot soil showed significant change at %1 probability level so that in the comparison test of Duncan means, the highest relative growth rate (0/138g) and the lowest relative growth rate of plant (0/073g) were observed, respectively.

Changes R / S (ratio of root dry weight to shoot dry weight)

By increasing the concentration of NaCl in the potting soil, R/S rate showed significant changes at %5 probability level so that in the comparison test of Duncan means, the highest R/S rate (0/173 g) and the lowest R/S rate of plant (0/101g) were observed, respectively.

DISCUSSION AND CONCLUSION

The results of the first test

The results of the germination rate

The results showed that with increasing salinity level, the rate and germination percentage is reduced, so that the highest germination rate in the control group and the lowest percentage of germination of 1 treatment (one-tenth percent concentration of sodium chloride) were observed, respectively. In ammonium nitrate treatment, it was also observed that with increasing concentration, the percent and germination rate are reduced. Generally salinity causes delayed germination and reduced percentage and germination rate. Depending on the severity of salinity, it can cause lack of or delay in seed germination and emergence and undermines the yield of plant production. Some plants such as wheat and barley have low tolerance to salinity in the seed germination stage, but their tolerance is increased at later growth stages [16]. Mean comparisons showed that the highest germination percentage (79%) was obtained in water with zero salinity. Research on Allerolfea occidentalis showed that with increasing concentration of NaCl, germination rate is stopped and germination rate is also reduced [17]. In addition to the potential of free water, salinity will also affect in the seed germination by toxic effects of ions such as Na^+ and Cl^-

[18]. Studies show that germination percentage and rate decrease with increasing salinity [19]. To start metabolic activity in the plants, it is necessary that the amount of water to be absorbed by them, which it is different depending on the chemical composition and permeability of seeds shell [20]. For each species, there is certain water potential that germination cannot occur in it [21]. According to the components of germination and vigor of plant, it can be concluded that the dill medicinal plant is sensitive to the salinity level in irrigation water more than four dS in term of germination [18]. With an increase in the salinity, percentage and rate of seed germination decrease sharply [22]. The results of the research are consistent with the above-mentioned results. The probable cause for reduced germination characteristics in different levels of salinity (sodium chloride) can be toxic effects of ions such as Na^+ and Cl^-. Reduced rate and germination percent in different concentrations of ammonium nitrate can be traced back to the inhibitory effect of ammonium for nitrate uptake.

Root and shoot

The results showed that the maximum length of root and shoot in the control group and the minimum length of root and shoot in Group A (with a concentration of 10% sodium chloride) were observed. As well, the maximum length of root and shoot in ammonium nitrate treatment in the control group and the minimum length of the treatment of (ammonium nitrate at a concentration of 10%) were observed, respectively. In a study of safflower cultivar, it was observed that root and shoot length as well as dry weight of root and shoot were decreased with increasing salinity levels in all cultivars [23]. In the research conducted on the species *Hedysarum criniferum* , it was observed that there are significant differences at 5% probability level in terms of the root and shoot length and vigor index at different levels of salinity. It also showed that with increasing salinity, root and shoot length and vigor were also decreased [24]. In addition, in the study of the seeds of *sisymbrium irio* (*Descurainia sophia*), the results indicated reduction in the percentage and germination rate, root and shoot length, root and shoot dry weight in different concentrations of salinity. Significant difference was observed between the different levels of salinity ($P \leq 0/01$). *Sisymbrium irio* was sensitive to high salinity in plant germination stage [25]. The results are consistent with the result of the above mentioned researchers. Reduced root and shoot growth by applying sodium chloride, is probably due to the toxic effects of ions such as Na^+ and $Cl^{-.}$ Yet the decreased root and shoot growth with treatment of ammonium nitrate may be due to changes in pH and ammonia toxic effect on the seed.

The results of the second test: Results of the effect of salinity and nitrogen on plant

Growth and Yield

The main stem and root length decreased with increasing salinity, however, by applying ammonium nitrate, it was gradually increased. Relative growth rate was also decreased with increasing salinity, yet with treatment of ammonium nitrate at concentrations up to 90 gr it was increased and declined with sodium chloride treatment at higher concentration of sodium chloride per 100 kg in the soil.

In addition, R/S ratio was reduced in different salinity levels and by adding ammonium nitrate; it was partly increased in salinity levels, so that no significant difference was observed in different levels of salinity.
It is generally recommended that fertilizer is a better choice for saline soils than normal conditions. Based on a general recommendation, if irrigation water salinity is more than 6 dS m, it is better to add 20 kg urea for every unit increase in the salinity. Urea fertilizers (urea derivatives) and urea with coated sulfur need the microbial activity and specific enzyme in the soil to be usable by grown plants, and since the microbial activity is very low in saline and sodic soils, the use of the fertilizer is not recommended in saline and sodic soils. As well, ammonium nitrate fertilizer is not recommended in saline and sodic for its high salinity and rapid dissolution and production a higher salinity than other nitrogen fertilizers [16]. Studies also show that adding nitrogen (in soils with nitrogen deficiency) showed that the addition of nitrogen, has not improved the growth and yield of a large number of plants, such as wheat, alfalfa, barley, beans, carrots, tomatoes, corn, clover, beans, millet while salinity was not too severe [26]. Considering that during the low nitrogen uptake for various reasons such as the permeability of plant roots, reduced soil microbial activity and subsequent reduction of mineralization of organic compounds, the absorption of nitrate is decreased in large amount in the root growth for the use of chlorine anion. In these conditions, nitrogen consumption could partly compensate for this problem and increase the yield. Also increased yield of wheat in saline conditions for further consumption of nitrogen fertilizer can be due to the reduction of sodium chloride concentration in plants [24]. By studying the interactional effects of salt and potassium nitrate fertilizer in the citrus seedlings in saline conditions, it was shown that uptake and nitrogen use yields were decreased. The results also showed that the optimal salinity increased nitrogen use in situations that shoot growth

temporarily affected by nitrogen absorption. The priority of nitrogen fertilizers in saline conditions is as follows: urea, ammonium sulfate, ammonium nitrate. Salinity index which is suggested as an indicator of salinity by fertilizers in ammonium nitrate, urea and ammonium sulfate is 104/7, 75/4 69 in per molecular weight of fertilizers, respectively [27]. Salinity stress with toxic effects of sodium and chlorine, and disturbance in the absorption of important elements, reduces the plant growth and yield, however, by applying ammonium nitrate to the plant prevents the absorption of sodium and chloride in plant growth and increases crop yield.

This study aimed to investigate the effect of salinity and nitrogen on the growth and morphological features of dill plant. For this purpose, various tests were carried out and the relevant findings were discussed. As mentioned at the beginning of the study, according to many properties of medicinal plants and humans need to use these drugs, their effective production is always of utmost importance. In short, it is hoped that by adopting appropriate strategies and policies based on a realistic understanding of the state of agriculture in the country and the use of correct scientific methods in all aspects, including planting, harvesting and industrial and economic exploitation, whether of nature or for mechanized cultivation, genuine understanding of the role and efficiency of medicinal plants growing in countries such as Iran will be reached and the protection of national assets to achieve prosperity and sustainable development of society will be accessible [28].

CONFLICT OF INTEREST
Authors declare no conflict of interest

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