The Effect of Drought Stress and Nitrogen Fertilizer Levels on Growth and Essential Oil of Savory (Satureja hortensis L.)

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ABSTRACT
In order to study the effect of drought stress and nitrogen fertilizer levels on growth and essential oil of savory (Satureja hortensis L.), this experiment was conducted in 2012 at Mahidasht area of Kermanshah province, Iran. The experiment was conducted in split plot in the form of a randomized complete block design with three replications. Treatments of the experiment included drought stress in three levels as the main plot (irrigation in every three, six and nine days) and nitrogen fertilizer in three levels as the sub plot (0, 100 and 200 kg N/ha). Results indicated that drought stress significantly affected plant height, the number of lateral branches, shoot fresh yield, shoot dry yield and essential oil yield; the highest value of all these traits was achieved when the field was irrigated every six days. Drought stress had no significant effect on essential oil percentage. Application of 100 kg N/ha resulted in the highest plant height, the number of lateral branches, shoot fresh yield, shoot dry yield and essential oil yield. However, the highest essential oil percentage was achieved when 200 kg N/ha was applied. The lowest value of all measured traits was observed in the control (0 kg N/ha). Results also showed that the interaction of drought stress × nitrogen fertilizer had no significant effect on any of the measured traits.

Keywords: Drought stresses, essential oil percentage, nitrogen, yield.

INTRODUCTION
Savory (Satureja hortensis L.) is an annual shrub plant; belonging to the Lamiaceae family [1]. The origin of this plant is eastern parts of the Mediterranean area [2]. The main compounds in the essential oil of savory are carvacrol, γ-Terpinene and p-Gymene [3]. Savory has carminative and mucus stimulation features and is used to cure respiratory system diseases, cough, anorexia, rheumatic pains, nervous pains, muscle cramps and pains, nausea, indigestion and diarrhea [4-6].

Low water availability in Iran has been always considered as one of the most important limiting factors for the production of crop and medicinal plants. The effect of drought stress on plants yield and the essential oil of medicinal plants has various aspects which requires overwhelming studies to be fully understood. It seems that the yield and essential oil of medicinal plants response differently to water shortage. To find these features and responses, complicated studies with various treatments on medicinal plants are required [7]. Jalilvand et al. [8] reported that drought stress reduced plant height and the number of lateral branches in savory. Charles et al. [9] found that dry weight and essential oil yield of mint increased when the field was irrigated more frequently. El-Mekawy [10] studied thyme plants and reported that irrigation interval of 10 days was better that 20 or 30 days in order to produce taller plants with more branches and higher fresh and dry weights.

Mineral nutrients such as nitrogen have important effects on plants vegetative and reproductive growth phases; affecting plant yield and the quality and quantity of the essential oil of the medicinal plants [11]. When available nitrogen is lower or higher than the plant required level, it disturbs the vital processes in plant body which appears in different ways such as increased, reduced or even stopped vegetative or reproductive growth. Because in medicinal plants the most important objective is the purity and healthiness of the essential oil, so attention must be paid to the application of the chemical fertilizers such
as nitrogen. It means than determining the accurate amount of chemical fertilizer is of a high importance [12]. Moumivand et al. [13] reported that application chemical nitrogen fertilizer had significant effect on plant height and dry weight of flowering shoots of savory. Alizadeh Sahzabi et al. [12] represented that application of 150 kg N/ha in soil and foliar application of 7.5% resulted in the highest essential oil yield, plant height and the number of lateral branches. Bist et al. [14] also found that increasing the application rate of chemical nitrogen fertilizer resulted in the enhancement of dill essential oil content. Regarding the increasing need of medication, food and cosmetic industries to medicinal plants as the primary materials, and the importance of the cultivation of these plants, the objective of this experiment was to evaluate the effect of drought stress and nitrogen fertilizer levels on growth and essential oil yield of savory.

**MATERIALS AND METHODS**

This experiment was conducted in 2012 at fields of the Agricultural Training Center of Kermanshah province, located in Mahidasht area of Kermanshah province, Iran (34° 16΄ N, 46° 49΄ E, 1365 m above the sea level). Mahidasht area, located in the south-west of Kermanshah, has semi-dry cold weather, with average annual precipitation of 335.1 mm and average daily temperature of 14.1°C. The experiment was conducted in split plot in the form of a randomized complete block design with three replications. Drought stress in three levels (irrigated every 3, 6 or 9 days) was in the main plots and chemical nitrogen fertilizer in three levels (0, 100 and 200 kg N/ha) was in the sub plots. Prior to the experiment, soil samples were taken from 0-30 cm depth in order to determine the physico-chemical properties of the test site soil. The soil was a loamy silt clay (clay, 36%; silt, 46%; sand, 18%) with the pH of 7.6 and EC of 0.8 ds/m. Other soil properties are listed in Table 1.

Savory seeds were planted at the depth of 1 cm below soil on May 4th. Irrigation was repeatedly conducted until the seedlings were established in the field; then, drought stress treatments were applied. Required amount of nitrogen fertilizer (urea) was applied in three parts: the first part was applied after germination phase, the second part was applied at the vegetative growth phase and the third part was applied before flowering. Prior to harvest, 20 plants were randomly selected from each plot to measure the desired traits. In order to eliminate the marginal effect, two side rows of each plot and 0.5 m from the both side of each row was removed. Then, the remaining area was used to obtain the yield. Flowering shoots were first dried under shadow and then by the means of a 50°C oven. After that, 100 g of the dried leaves were grinded and the essential oil was produced using a cleverenger in 3 h. The measured traits included: plant height, the number of lateral branches, plant fresh and dry weight, shoot fresh and dry yield, essential oil percentage and essential oil yield. Finally, data were analyzed using SAS and means were compared according to the Duncan’s multiple range test at P≤0.05.

**RESULTS**

**Plant height**

Analysis of variance showed that the effect of drought stress was significant on plant height at P≤0.01 (Table 2). The highest plant height (66.07 cm) was achieved when the field was irrigated every six days and the lowest plant height (58.67 cm) was achieved when the field was irrigated every nine days (Fig. 1). Results indicated that nitrogen levels had significant effect on savory plant height at P≤0.01 (Table 2). Plant height was the highest (65.13 cm) in 100 kg N/ha treatment, without any significant differences from 200 kg N/ha. The lowest plant height (59.44 cm) was achieved in the control (0 kg N/ha) (Fig. 2).

**The number of lateral branches**

Analysis of variance indicated the significant effect of drought stress on the number of lateral branches of savory plants at P≤0.01 (Table 2). The highest number of branches (26.09) was related to the irrigation in every six days and the lowest number of lateral branches (23.27) was related to the irrigation in every nine days (Fig. 3).

Analysis of variance also showed that nitrogen level had significant effect on the number of lateral branches at P≤0.01 (Table 2). The highest number of lateral branches (25.65) was achieved when 100 kg N/ha was applied and the lowest number of lateral branches (22.79) was achieved in the control. There was no significant differences between 100 and 200 kg N/ha (Fig. 4).

**Shoot fresh yield**

Analysis of variance indicated the significant effect of drought stress on savory shoot fresh yield at P≤0.01 (Table 2). The highest shoot fresh yield (5.96 kg/m²) was achieved when the field was irrigated every six days and the lowest shoot fresh yield (4.62 kg/m²) was achieved when the field was irrigated every nine days (Fig. 5).
Results also indicated that nitrogen levels had significant effect on shoot fresh yield at P≤0.01 (Table 2). The highest shoot fresh yield (5.87 kg/m²) was achieved in 100 kg N/ha which was significantly the same as 200 kg N/ha. The lowest shoot fresh yield (4.46 kg/m²) was achieved in the control (Fig. 6).

**Shoot dry yield**

Analysis of variance indicated that drought stress had significant effect on shoot dry yield at P≤0.01 (Table 2). The highest shoot dry yield (1.55 kg/m²) was achieved when the field was irrigated every six days and the lowest shoot dry yield (1.16 kg/m²) was achieved when the field was irrigated every nine days (Fig. 7).

Analysis of variance also showed that nitrogen level had also a significant effect on shoot dry yield (Table 2). Shoot dry yield was the highest (1.46 kg/m²) in 100 kg N/ha and the lowest (1.17 kg/m²) in the control (Fig. 8).

**Essential oil percentage**

Results represented that drought stress had no significant effect on essential oil percentage; however, nitrogen levels had significant effect on this trait (Table 2). Mean comparison showed that the highest essential oil percentage (1.68%) was achieved when 200 kg N/ha was applied and the lowest essential oil percentage (1.51%) was achieved in the control in which no chemical nitrogen was applied (Fig. 9).

**Essential oil yield**

Analysis of variance showed that drought stress significantly affected savory essential oil yield (Table 2). The highest essential oil yield (9.77 g/m²) was achieved when the field was irrigated every six days and the lowest essential oil yield (8.21 g/m²) was achieved when the field was irrigated every nine days (Fig. 10).

Analysis of variance also indicated that nitrogen levels had significant effect on essential oil yield at P≤0.01 (Table 2). The highest essential oil yield (9.68 g/m²) was achieved in 100 kg N/ha which was significantly the same as 200 kg N/ha. The lowest essential oil yield (7.63 g/m²) was achieved in the control (Fig. 11).

Table 1. Physico-chemical properties of the test site soil.

<table>
<thead>
<tr>
<th>OC (%)</th>
<th>N (%)</th>
<th>P (mg/kg)</th>
<th>K (mg/kg)</th>
<th>Mn (mg/kg)</th>
<th>Fe (mg/kg)</th>
<th>Zn (mg/kg)</th>
<th>Cu (mg/kg)</th>
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<tr>
<td>1.0</td>
<td>0.1</td>
<td>20</td>
<td>230</td>
<td>5.8</td>
<td>8.2</td>
<td>1.14</td>
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Table 2. Analysis of variance of the effect of treatments on the measured traits.

<table>
<thead>
<tr>
<th>SOV</th>
<th>df</th>
<th>Plant height</th>
<th>The number of lateral branches</th>
<th>Shoot fresh yield</th>
<th>Shoot dry yield</th>
<th>Essential oil percentage</th>
<th>Essential oil yield</th>
</tr>
</thead>
<tbody>
<tr>
<td>Rep</td>
<td>2</td>
<td>*</td>
<td>**</td>
<td>**</td>
<td>**</td>
<td>Ns</td>
<td>*</td>
</tr>
<tr>
<td>Drought stress</td>
<td>2</td>
<td>**</td>
<td>**</td>
<td>**</td>
<td>**</td>
<td>Ns</td>
<td>*</td>
</tr>
<tr>
<td>Error</td>
<td>4</td>
<td>19.13</td>
<td>3.76</td>
<td>130.83</td>
<td>11.12</td>
<td>0.042</td>
<td>0.09</td>
</tr>
<tr>
<td>N level</td>
<td>2</td>
<td>*</td>
<td>**</td>
<td>**</td>
<td>**</td>
<td>*</td>
<td>*</td>
</tr>
<tr>
<td>Drought × N</td>
<td>4</td>
<td>ns</td>
<td>ns</td>
<td>ns</td>
<td>ns</td>
<td>ns</td>
<td>ns</td>
</tr>
<tr>
<td>Error</td>
<td>39</td>
<td>9.23</td>
<td>1.02</td>
<td>74.60</td>
<td>4.31</td>
<td>0.04</td>
<td>0.94</td>
</tr>
</tbody>
</table>

CV (%) - 4.84  4.11  16.24  15.73  12.07  10.95

ns, nonsignificant; *, significant at P≤0.05; **, significant at P≤0.01.

Figure 1. The effect of drought stress on plant height.
Figure 2. The effect of nitrogen levels on plant height.

Figure 3. The effect of drought stress on the number of lateral branches.

Figure 4. The effect of nitrogen levels on the number of lateral branches.

Figure 5. The effect of drought stress on shoot fresh yield.
Figure 6. The effect of nitrogen levels on shoot fresh yield.

Figure 7. The effect of drought stress on shoot dry yield.

Figure 8. The effect of nitrogen levels on shoot dry yield.

Figure 9. The effect of nitrogen levels on essential oil percentage.
DISCUSSION
Results indicated that by enhancing the severity of drought stress, changes of plant height, the number of lateral branches, shoot fresh and dry yield and essential oil yield did not followed a regular trend in the way that by increasing the irrigation interval from three to six days, the value of the measured increased; however, by increasing the irrigation interval from six to nine days, the values decreased. Briefly, the measured traits reached their highest values in six day irrigation and their lowest values in nine days irrigation intervals.

Water shortage is a limiting factor to agricultural production. So, drought stress induced by increasing the interval of irrigations resulted in the reduction of plant growth and development. Results of our experiment showed that irrigating the field every nine days resulted in the reduction of the measured traits; these findings are in agreement with the results of other experiments [15-18]. Results of this experiment also indicated that nitrogen levels had also significant effect on the measured traits; application of 100 kg N/ha resulted in the highest plant height, the number of lateral branches, shoot fresh and dry yield and essential oil yield. This amount of nitrogen fertilizer had the highest effect on the vegetative phase of savory. However, the highest essential oil percentage was achieved when 200 kg N/ha was applied.

Nitrogen is the most important nutrient for the synthesis of proteins and the enhancement of nitrogen content in plants, up to a certain level, results in the enhancement of plant protein content, consequently increasing plants vegetative growth [19]. Nitrogen is one of the nutrients which is required in all stages of plants growth. Enhancement of essential oil yield as the result of nitrogen application in our experiment may be attributed to the role of nitrogen in division and development of essential oil containing cells, essential oil tubes secretory canals and secretory trichomes. These results are in agreement with the findings of other experiments [12, 13, 14, 20].

CONCLUSION
Regarding the results of this experiment, irrigating the field every six days and applying 100 kg N/ha had the highest improving effect on savory growth and essential oil yield.
REFERENCES


Citation of This Article