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Effects of zinc and nitrogen application on agronomic traits and qualitative characteristic of sunflower in saline condition

Mohammad Taher Nezami* and Ghazaleh Vafaei

Department of Soil Science, Islamic Azad University, Karaj Branch, Karaj, Iran.

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This present study was performed at the Department of Soil Science of Karaj Branch, during the two seasons of 2008 to 2009 and 2009 to 2010. The experimental design aimed to study the response of sunflower to different nitrogen and zinc fertilizer levels under two doses of salinity stress. The obtained results from the two years showed that nitrogen increased seed yield while there was no significant differences between the two nitrogen levels; also, results showed that zinc application had not significant effect on seed yield. Conversely, salinity decreased seed yield and yield components dramatically. Based on mean comparison, plant height increased on account of nitrogen and zinc application. Nitrogen fertilizer application improved seed number per silique and 1000-seed weight over two years. Among yield components only number of seed per silique was affected by zinc in the first year. Oil percentage decreased due to nitrogen, zinc and salinity in the first year but zinc had no significant effect on oil in the second year. Salinity stress increased glucosinolate and protein content. Other treatments had no significant effect on these traits. Salt stress induction decreased N, P, K, Ca and Mg content while increased Na, Cl and Na/K ratio.

Key words: Sunflower, nitrogen, salinity, yield and yield components, zinc.

INTRODUCTION

Salt stress is a major abiotic stress reducing the productivity of crops in many areas of the world (Yamaguchi and Blumwald, 2005). This is particularly the case in semi-arid and arid zones, where already over a decade ago 50% of the cropland was salt affected (Flower and Yeo, 1995). Soil salinity occurs from natural processes or from crop irrigation with saline water (Hasegawa et al., 2000). Salinity affects the water balance and results in osmotic damage; however, osmotic adjustment is a plant adaptation mechanism used to maintain their water balance in plant (Sairam and Tyagi, 2004). This can lead to a reduction of biodiversity and land degradation (Ghassemi et al., 1995). In many plant species, soil salinity is known to reduce growth and development through osmotic stress, ion toxicity, mineral deficiencies and induced physiological and biochemical disorders in metabolic processes (Hasegawa et al., 2000). Salinity stress is often associated with nutritional imbalance. The interaction between salt stress and other environmental factors influence the plant's response to the stress (Ashraf and McNeilly, 2004). Nitrogen and Zinc are essential minerals required for normal physiological processes of plants. Nitrogen fertilizer is the most important element for crop growth and high yield with good quality where, it causes an increase in photosynthesis rate, metabolites synthesized, meristematic activity and assimilates transport to the seed. Seed yield and yield attributes increased by increasing nitrogen levels (Ali and Zaman, 1997). Nitrogen fertilizer increases yield by influencing a variety of growth parameters such as the number of branches per plant, the number of pods per plant, the total plant weight, the leaf area index (Henry and McDonald, 1978). Also, it increases the number and weight of pods, seeds and flowers per plant, and overall crop assimilation, contributing to increased seed yield (Al-Barrak, 2006).

*Corresponding author. E-mail: Taher.nezami@yahoo.com.
Zinc is an essential microelement required by higher plants, and is mainly absorbed in the form of Zn\(^{2+}\). Zinc also plays an important role in the production of biomass (Cakmak, 2008). In addition, Zn plays other indirect and significant roles as stabilizer of proteins, membranes, and DNA-binding proteins such as Zn-fingers (Aravind and Prasad, 2003). Furthermore, zinc may be required for chlorophyll production, pollen function and fertilisation (Pandey et al., 2006). Zinc is required for the biosynthesis of the plant growth regulator such as indole-3-acetic acid (IAA) (Fang et al., 2008), and for carbohydrate and N metabolism which leads to high yield and yield components.

Zinc is an essential micronutrient for higher plants especially oil crops where it is required for the activity of various types of enzymes (dehydrogenases, RNA and DNA polymerases), carbohydrate metabolism and protein synthesis. Zinc also plays an important role in the production of biomass (Kaya and Higgs, 2002). Furthermore, zinc may be required for chlorophyll production, pollen function and fertilization (Pandey et al., 2006). Zinc deficiency also affects carbohydrate metabolism, damages pollen structure, and decreases the yield (Fang et al., 2008). Bybordi and Malakouti (2007) found that application of zinc had a significant effect on seed yield, seed oil content and 1000-seed weight. Zinc deficiency is one of the most widespread micronutrient deficiencies in Iran as a result for the alkaline soil condition. So, it is very important to apply zinc fertilizer for increasing crop yields and improving crop quality. The family Brassicaceae includes a number of species that have considerable nutritional and economic values and that have been under cultivation since 1500 B.C. These crops are extensively grown as cash crops, fodder and industrial/medicinal crops (Ashraf and McNeilly, 2004). The most common Brassica oilseed crops grown in the world for industrial purpose are rape-seeds, Brassica campestris and Brassica napus. Therefore, this investigation aimed to study the response of sunflower to nitrogen and zinc fertilizer under conditions of salt stress.

**MATERIALS AND METHODS**

Two pot experiments were conducted at the Department of soil science, Karaj branch, Islamic Azad University, Karaj, Iran, during the two seasons of 2008 to 2009 and 2009 to 2010 to study the response of sunflower to different nitrogen and zinc fertilizer levels under two doses of salinity stress. The experiment was conducted in a glasshouse, in plastic pots (30 cm diameter and 40 cm depth) containing 20 kg soil. The soil was sandy loam, moderately calcareous, low in nitrogen, low in organic matter and alkaline in reaction having a pH of 7.7. Treatments were consisting of different nitrogen levels (0, 50 and 100 kg ha\(^{-1}\) nitrogen from urea), different zinc levels (0, 5 and 10 kg ha\(^{-1}\) zinc from zinc sulfate) and two dose of saline water (8 and 16 dS m\(^{-1}\)). The amount of urea and zinc sulfate was calculated based on pot surface. All the pots were fertilized with above mentioned fertilizers so that fertilizers were incorporated into the soil before seeding. The pots were arranged according to three-factor randomized complete block design (RCBD) in three replications. The growth conditions were as follows: photoperiod of 14/10 h (day/night), temperature of 25/18°C (day/night), and the maximal photosynthetic photon flux density of 600 \(\mu\)mol m\(^{-2}\) s\(^{-1}\). The pots were labelled and randomly arranged in the greenhouse and rearranged several times during the growth period. The seed surface was sterilized by immersion in 2% sodium hypochloride solution for 10 min and 96% ethanol for 30 s, then seeds were washed with distilled water for several times. Ten seeds were sown directly in plastic pots. Immediately after sowing, soils were watered and watering was carried out regularly every two days during the experiment. The sunflower seedlings were thinned to four uniform stands two weeks after planting. After one weeks of sowing, the salt treatments were applied. Salt treatments were applied by adding appropriate amount of NaCl in irrigation water. The EC of the irrigation water was 8 and 16 dS m\(^{-1}\). To avoid early plant death by a sudden salt stress shock of the young seedlings, the salt stress was imposed gradually by applying half of the salt concentration over two weeks and increasing it to the final concentration for the rest of the plants. The pots were watered and watering was carried out regularly every two days. The salt treatments were maintained until final harvest. At harvest time, plants were sampled to estimate, plant height, and number of silique per plant, number of seeds per silique, 1000-seed weight, and seed yield per plant. Seed oil percentage was analyzed according to A.O.A.C. (1980) with Soxhelt apparatus. Glucosinolate was extracted and analysed as previously described with minor modifications (Jia et al., 2009). Samples were boiled in 4 ml water for 10 min. After recovery of the liquid, the residues were washed with water, and the combined aqueous extract was applied to a DEAE-Sephadex A-25 column (pyridine acetate form). The column was washed three times with 20 mM pyridine acetate and twice with water. The glucosinolates were converted into their desulpho analogues by overnight treatment with 100 \(\mu\)l of 0.1% (1.4 units) aryl sulphatase, and the desulphoglucosinolates were eluted with 2 \(\times\) 0.5 ml water. HPLC analysis was performed using an HPLC system consisting of a Waters 2695 separations module and a Waters 2996 photodiode array detector (Waters Corp., Milford, MA, USA). The HPLC system was connected to a computer with Empower Pro software. A Hypersil C18 column (5 lm particle size, 4.6 mm 250 mm; Elite Analytical Instruments Co. Ltd., Dalian, China) was used with a mobile phase of acetonitrile and water. The flow rate was 1 ml min\(^{-1}\) for 15 min followed by isocratic elution with 20% acetonitrile over the next 15 min followed by isocratic elution with 20% acetonitrile for the final 10 min. A 40-\(\mu\)l sample was injected onto the column by an auto sampler. Absorbance was detected at 226 nm. Singing was used as an internal standard for HPLC analysis. Desulphoglucosinolates were identified by comparison of retention time and quantified by peak area. For calculation of molar concentrations of individual glucosinolates, the relative response factors reported by Brown Tokuhisa et al. (2003) were used to correct for absorbance differences between the standard and the other glucosinolates. The glucosinolate concentration was expressed as ng/g dry weight (DW). Protein percentage was estimated by using Inframatic 8620 Piercer.

Total N and P were determined through titration method by Kjeltac Auto 1030 Analyzer, Tecator and calorimetric method by Spectrophotometer, 6505 JenWay, respectively. For ion determination, fresh samples were extracted in 0.1 N nitric acid. Na\(^{+}\), Mg\(^{2+}\), and Ca\(^{2+}\) contents in the samples were detected by atomic absorption. K\(^{+}\) was detected by flame photometry in the samples from sunflower plants. For chloride determination, Cl\(^{-}\) was determined by the silver ion-titration method with an automatic chloridometer (Buckheimer-Cotlove chloridometer) according to Bozok (1970).

All data were analyzed from analysis of variance (ANOVA) using the SAS. Duncan’s Multiple Range Test was used to measure statistical differences between treatments. Also the charts were drawn by Excel.
RESULTS AND DISCUSSION

The analysis of variance showed that the application of nitrogen had significant effect on seed yield, plant height, seed number per silique, 1000 seed weight, oil percentage and seed magnesium content in the first year (Table 1), while in second year, effect of nitrogen fertilizer was significant only on seed yield, plant height, seed number per silique, 1000 seed weight, oil percentage and seed magnesium content (Table 2). Regarding zinc application, we observed that there were significant effects on plant height, seed number per silique and oil percentage in the first year, and on seed nitrogen content in the second year (Tables 1 and 2). In case of salt stress, results indicated that salinity had significant effect on all traits except for seed number per silique, oil...
percentage, glucosinolate, phosphorus and magnesium content in the first year (Table 1). In the second year, salinity showed no significant effect on glucosinolate, phosphorus, calcium, magnesium and sodium to potassium ratio (Table 2). In some traits interactions were significant which are illustrated in following the figures.

### Seed yield

Comparison of means demonstrated that nitrogen application increased seed yield in both years, although there was no significant difference between two doses of nitrogen (Tables 3 and 4). In addition, increase of salinity up to 16 dS.m⁻¹ significantly decreased seed yield over two years. Increased seed yields due to nitrogen application were also reported by Nourai (1982). The increase in yield and yield attributes may be due to nitrogen fertilizer that caused an increase in photosynthesis rate, metabolites synthesized and assimilates transport from source to the sink.

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**Table 3.** Mains effects of nitrogen, zinc and salinity on sunflower traits (First year).

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Level</th>
<th>Yield</th>
<th>Height</th>
<th>Silique per plant</th>
<th>Seed per silique</th>
<th>Seed weight</th>
<th>Oil</th>
<th>Glucosinolate</th>
<th>Protein</th>
<th>N</th>
<th>P</th>
<th>K</th>
<th>Ca</th>
<th>Mg</th>
<th>Na</th>
<th>Cl</th>
<th>Na/K</th>
</tr>
</thead>
<tbody>
<tr>
<td>Nitrogen</td>
<td>0 kg</td>
<td>59.38</td>
<td>41.88</td>
<td>11.86</td>
<td>23.83</td>
<td>2.29</td>
<td>41.55</td>
<td>28.83</td>
<td>11.86</td>
<td>1.36</td>
<td>0.12</td>
<td>2.42</td>
<td>1.31</td>
<td>0.33</td>
<td>9.90</td>
<td>10.83</td>
<td>4.26</td>
</tr>
<tr>
<td></td>
<td>50 kg</td>
<td>131.88</td>
<td>93.55</td>
<td>11.94</td>
<td>35.55</td>
<td>2.70</td>
<td>41.11</td>
<td>27.00</td>
<td>11.94</td>
<td>2.25</td>
<td>0.12</td>
<td>2.43</td>
<td>1.34</td>
<td>0.33</td>
<td>8.95</td>
<td>10.44</td>
<td>3.78</td>
</tr>
<tr>
<td></td>
<td>100 kg</td>
<td>138.72</td>
<td>99.05</td>
<td>11.88</td>
<td>41.00</td>
<td>2.90</td>
<td>40.44</td>
<td>27.38</td>
<td>11.88</td>
<td>2.66</td>
<td>0.11</td>
<td>2.38</td>
<td>1.28</td>
<td>0.34</td>
<td>9.17</td>
<td>10.30</td>
<td>3.97</td>
</tr>
<tr>
<td>Zinc</td>
<td>0 kg</td>
<td>100.44</td>
<td>75.83</td>
<td>11.96</td>
<td>31.27</td>
<td>2.61</td>
<td>41.61</td>
<td>27.83</td>
<td>11.96</td>
<td>2.10</td>
<td>0.11</td>
<td>2.37</td>
<td>1.28</td>
<td>0.34</td>
<td>9.15</td>
<td>10.22</td>
<td>3.99</td>
</tr>
<tr>
<td></td>
<td>5 kg</td>
<td>113.61</td>
<td>78.16</td>
<td>11.87</td>
<td>33.50</td>
<td>2.60</td>
<td>40.88</td>
<td>28.27</td>
<td>11.87</td>
<td>2.08</td>
<td>0.12</td>
<td>2.41</td>
<td>1.32</td>
<td>0.33</td>
<td>9.44</td>
<td>11.00</td>
<td>4.04</td>
</tr>
<tr>
<td></td>
<td>10 kg</td>
<td>115.94</td>
<td>80.50</td>
<td>11.85</td>
<td>35.16</td>
<td>2.68</td>
<td>40.61</td>
<td>27.11</td>
<td>11.85</td>
<td>2.08</td>
<td>0.12</td>
<td>2.45</td>
<td>1.32</td>
<td>0.33</td>
<td>9.44</td>
<td>10.36</td>
<td>3.98</td>
</tr>
<tr>
<td>Salinity</td>
<td>8 dS/m</td>
<td>137.14</td>
<td>102.96</td>
<td>12.45</td>
<td>32.70</td>
<td>2.88</td>
<td>40.88</td>
<td>27.00</td>
<td>12.45</td>
<td>2.14</td>
<td>1.45</td>
<td>2.54</td>
<td>1.35</td>
<td>0.33</td>
<td>4.11</td>
<td>6.79</td>
<td>1.62</td>
</tr>
<tr>
<td></td>
<td>16 dS/m</td>
<td>82.85</td>
<td>53.37</td>
<td>11.34</td>
<td>28.92</td>
<td>2.37</td>
<td>37.18</td>
<td>29.48</td>
<td>11.34</td>
<td>2.03</td>
<td>1.14</td>
<td>2.28</td>
<td>1.27</td>
<td>0.34</td>
<td>14.57</td>
<td>14.25</td>
<td>6.39</td>
</tr>
</tbody>
</table>

Values within the same column and followed by the same letter are not significantly different at $P < 0.05$ by an ANOVA protected Duncan’s Multiple Range Test.

**Table 4.** Mains effects of nitrogen, zinc and salinity on sunflower traits (Second year).

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Level</th>
<th>Yield</th>
<th>Height</th>
<th>Silique per plant</th>
<th>Seed per silique</th>
<th>Seed weight</th>
<th>Oil</th>
<th>Glucosinolate</th>
<th>Protein</th>
<th>N</th>
<th>P</th>
<th>K</th>
<th>Ca</th>
<th>Mg</th>
<th>Na</th>
<th>Cl</th>
<th>Na/K</th>
</tr>
</thead>
<tbody>
<tr>
<td>Nitrogen</td>
<td>0 kg</td>
<td>56.38</td>
<td>37.88</td>
<td>12.46</td>
<td>34.83</td>
<td>1.16</td>
<td>41.55</td>
<td>33.83</td>
<td>12.46</td>
<td>2.23</td>
<td>0.31</td>
<td>3.20</td>
<td>1.45</td>
<td>0.34</td>
<td>27.33</td>
<td>17.66</td>
<td>8.54</td>
</tr>
<tr>
<td></td>
<td>50 kg</td>
<td>128.88</td>
<td>89.55</td>
<td>12.64</td>
<td>36.44</td>
<td>1.66</td>
<td>39.22</td>
<td>35.27</td>
<td>12.64</td>
<td>2.28</td>
<td>0.31</td>
<td>3.25</td>
<td>1.46</td>
<td>0.33</td>
<td>28.44</td>
<td>20.66</td>
<td>8.28</td>
</tr>
<tr>
<td></td>
<td>100 kg</td>
<td>135.72</td>
<td>95.05</td>
<td>12.63</td>
<td>39.00</td>
<td>2.64</td>
<td>39.16</td>
<td>34.83</td>
<td>12.63</td>
<td>2.43</td>
<td>0.32</td>
<td>3.31</td>
<td>1.42</td>
<td>0.33</td>
<td>28.05</td>
<td>19.61</td>
<td>8.51</td>
</tr>
<tr>
<td>Zinc</td>
<td>0 kg</td>
<td>97.44</td>
<td>71.83</td>
<td>12.65</td>
<td>37.00</td>
<td>1.73</td>
<td>40.00</td>
<td>34.66</td>
<td>12.65</td>
<td>2.31</td>
<td>0.31</td>
<td>3.20</td>
<td>1.47</td>
<td>0.34</td>
<td>26.50</td>
<td>19.16</td>
<td>8.36</td>
</tr>
<tr>
<td></td>
<td>5 kg</td>
<td>110.61</td>
<td>74.16</td>
<td>12.56</td>
<td>36.66</td>
<td>1.64</td>
<td>39.77</td>
<td>34.50</td>
<td>12.56</td>
<td>2.29</td>
<td>0.32</td>
<td>3.24</td>
<td>1.44</td>
<td>0.34</td>
<td>27.94</td>
<td>19.11</td>
<td>8.68</td>
</tr>
<tr>
<td></td>
<td>10 kg</td>
<td>112.94</td>
<td>76.50</td>
<td>12.53</td>
<td>36.31</td>
<td>1.68</td>
<td>40.16</td>
<td>34.77</td>
<td>12.53</td>
<td>2.34</td>
<td>0.31</td>
<td>3.32</td>
<td>1.42</td>
<td>0.34</td>
<td>27.38</td>
<td>19.66</td>
<td>8.28</td>
</tr>
<tr>
<td>Salinity</td>
<td>8 dS/m</td>
<td>134.14</td>
<td>98.96</td>
<td>12.85</td>
<td>39.48</td>
<td>1.81</td>
<td>41.37</td>
<td>33.51</td>
<td>12.85</td>
<td>2.45</td>
<td>0.32</td>
<td>3.41</td>
<td>1.45</td>
<td>0.34</td>
<td>25.74</td>
<td>15.40</td>
<td>8.40</td>
</tr>
<tr>
<td></td>
<td>16 dS/m</td>
<td>79.85</td>
<td>49.37</td>
<td>12.31</td>
<td>34.03</td>
<td>1.56</td>
<td>38.59</td>
<td>34.77</td>
<td>12.31</td>
<td>2.18</td>
<td>0.31</td>
<td>3.09</td>
<td>1.34</td>
<td>0.34</td>
<td>28.81</td>
<td>23.22</td>
<td>8.49</td>
</tr>
</tbody>
</table>

Values within the same column and followed by the same letter are not different at $P < 0.05$ by an ANOVA protected Duncan’s Multiple Range Test.
consequently increase of yield. In addition, El Hilo et al. (1970) reported that the lack of nitrogen induced yellowing of leaves and flower stalks and caused burning of leaf tips. The results (Tables 3 and 4) show that NaCl adversely affected the seed yield. The salt stress caused significant reductions in all the growth variables including dry weights (data are not shown). Salinity has both osmotic and specific ion effects on plant growth (Dionisio-Sese and Tobita, 2000). In this study, salt stress caused a significant decrease in the plant height, dry weights of root, shoot and leaf of sunflower (data are not shown) followed by decrease in seed yield. Reduction in plant growth as a result of salt stress has also been reported in several other plant species (Ashraf and O’leary, 1997; Turkmen et al., 2008).

**Plant height**

Means of plant height under the nitrogen fertilizer rates in both years showed an increase in plant height as nitrogen fertilizer rate increased (Tables 3 and 4). The previous results might be due to the positive effect of nitrogen on the growth development of stem and leaf, which was reflected into taller plants. By contrast, salinity mitigated plant growth. Similar results were found in both years. The most common adverse effect of salinity on the crop of *Brassica* is the reduction in plant height, size and yield as well as deterioration of the quality of the product (Kumar, 1995). Our results are also in accordance with previous finding of Viégas et al. (2001) who reported that salinity inhibits shoot growth. On the other hand, zinc improved plant growth and increased plant height. Zinc plays an important role in tryptophan biosynthesis, later is precursor of auxine so; it is expecting that zinc will increase plant growth and elongation. It has been reported that zinc played an important role in the production of biomass (Kaya and Higgs, 2002). Welch et al. (1982) stated that zinc is necessary for root cell membrane integrity. As suggested by Marschner and Cakmak (1986), root cell membrane permeability increased under zinc deficiency which might be related to the functions of zinc in cell membranes so, external zinc application could increased the integrity of cell membrane and play important role in plant enzyme systemss which may increase the assimilates. Also zinc is found in phosphoenolpyruvate carboxylase structure.

**Number of silique per plant**

As for the number of silique per plant, the mean values in Tables 3 and 4 revealed that the lowest value was produced from the plants treated with 16 dS.m⁻¹ saline water. According to the findings of Lin et al. (2004), salinity stress lead to abscisic acid synthesis and transport to shoots and finally abscission of flowers and siliqua. No significant difference was shown between the nitrogen and zinc rates in the number of silique per plant. It can be concluded that the similarity in silique number of different nitrogen and zinc rates may be attributed to their genetic spetotial. The number of silique branches per plant is the result of combined effect of genetic make up of the crop and environmental conditions, which plays a remarkable role towards the final seed yield of the crop (Sana et al., 2003).

**Seed number per silique**

Seed number per silique was affected by nitrogen, zinc
Figure 2. Interaction between different nitrogen levels and salinity stress in the first year showed that plant height was affected by these treatments so that increase of salinity level decreased plant height under each level of nitrogen fertilizer. Application of 100 kg.ha\(^{-1}\) nitrogen significantly increased plant height whether under 8 dS.m\(^{-1}\) or 16 dS.m\(^{-1}\) salinity stress.

Figure 3. Interaction between different nitrogen levels and salinity stress in the first year showed that seed weight was affected by these treatments so that increase of salinity level decreased sunflower seed weight under each level of nitrogen fertilizer. Application of 100 kg.ha\(^{-1}\) nitrogen significantly increased seed weight under conditions of 8 dS.m\(^{-1}\) salinity stress while application of this amount of nitrogen under conditions of 16 dS.m\(^{-1}\) salinity stress had not significant effect on seed weight.

and salinity stress in the first year, while in the second year zinc had no significant effect on seed number per silique. Seed number was increased along with increasing of nitrogen or zinc rate. Nitrogen fertilizer increases yield by influencing a variety of growth parameters such as the number of branches per plant, the number of pods per plant, the total plant weight, the leaf area index. Also, it increases the number and weight of seeds (Al-Barrak, 2006). One of the most important reasons for decreasing seed number per silique is that salinity decreases length of siliques. Sakr and assistants (2007), reported that seed number per silique is sensitive to salinity stress. In this regard, Mendham and Salibury (1995), showed that ovum number per silique is about 30 at seed set stage but decreased due to osmotic and other environmental stresses.
Interaction between nitrogen and zinc

Figure 4. Interaction between different nitrogen and zinc levels in the first year showed that zinc application deceased oil percentage under nitrogen free treatments. Nitrogen application increased oil percentage. The highest oil percentage was obtained when plants were treated by 50 kg ha\(^{-1}\) nitrogen and no zinc application, although there was no significant difference between 50 and 100 kg ha\(^{-1}\) nitrogen levels.

Interaction between nitrogen and zinc

Figure 5. Interaction between different nitrogen and zinc levels in the first year showed that zinc application deceased phosphorus content under nitrogen free treatments because of competitiveness effect between zinc and phosphorus while nitrogen application increased phosphorus accumulation.

1000 seed weight

In both years, nitrogen rate increased 1000-seed weight increased, while salinity decreased it. 1000 seed weight did not change due to zinc application. The lowest 1000-seed weight was obtained when salt stress occurred or no nitrogen was applied which can be attributed to the deficiency of resources (photosynthesis from lower number of leaves and pods) to support pod filling (Angadi et al., 2003). Olsson (1960) found that 1000-seed weight was not strongly or little influenced by environmental conditions, respectively. However, Krogman and Hobbs (1975) concluded that 1000-seed weight was increased with irrigation and nitrogen levels.

Oil percentage

As for oil percentage, the data of Tables 3 and 4 revealed that nitrogen free treatments had the highest and then
Figure 6. Interaction between different nitrogen levels and salinity stress in the second year showed that seed yield was strongly affected by these treatments so that increase of salinity level decreased seed yield under each level of nitrogen fertilizer. In addition, there was no significant different between 50 and 100 kg.ha\(^{-1}\) nitrogen.

Figure 7. Interaction between different nitrogen levels and salinity stress in the second year showed that plant height was affected by these treatments so that increase of salinity level decreased plant height under each level of nitrogen fertilizer. Application of 100 kg.ha\(^{-1}\) nitrogen significantly increased plant height whether under 8 dS.m\(^{-1}\) or 16 dS.m\(^{-1}\) salinity stress.

significantly decreased under the higher nitrogen rates. Similarly, salinity stress decreased oil percentage. Obtained results from zinc application differ from year to year in other words, zinc decreased oil content in the first year while had no significant effect on oil content in the second year (Tables 3 and 4). Several reasons have been given for the decrease in oil content with increasing nitrogen rates. This may be due to a better supply of nitrogen increase on the formation of nitrogen containing protein precursors so that protein formation competes more strongly for photosynthates; as a result less of the latter are available for fat synthesis Holmes (1980). Kutcher et al. (2005) stated that it might be due to the dilution effect of increased seed yield with increased nitrogen fertilization and the inverse relationship of protein and oil content. These results are in harmony with Ahmadi and Bahrani (2009).

**Glucosinolate**

Nitrogen and zinc had no significant effect on glucosinolate accumulation in seed over two years while salt stress significantly increased glucosinolate content.
Figure 8. Interaction between different nitrogen and zinc levels in the second year showed that nitrogen and zinc increased phosphorus content compared with the control treatment although there was no significant difference among treatments.

Figure 9. Interaction between different nitrogen and zinc levels in the second year showed that potassium content increased as result of increase of zinc in those plants which were treated by 50 kg ha\(^{-1}\) nitrogen. When 100 kg ha\(^{-1}\) nitrogen was applied zinc had not positive effect on potassium accumulation.

(Tables 3 and 4). Little is known about glucosinolates accumulation in response to salt stress, although previous studies indicate that environmental factors such as light (Engelen-Eigles et al., 2006), temperature (Velasco et al., 2007) and heavy metals (Tolra et al., 2006) alter the glucosinolate content and composition. Recently, several studies showed that salt stress dramatically increased the total glucosinolate content in broccoli florets (Lopez-Berenguer et al., 2008, 2009); suggesting that they could be involved in the osmotic adjustment under low water potential (Qasim et al., 2003).

Protein content

The increasing of nitrogen fertilizer rate might increase the absorbed nitrogen by plant root. Accordingly, nitrogen increases in protein metabolism and is reflected in increasing the protein content in sunflower seeds. Conversely, our results show that increased nitrogen levels had no significant effect on seed protein content (Tables 3 and 4). Our findings are consistent with the results of Singh (2000), which suggest that protein content was not affected by fertilizers treatment and is related to genetic potential. Similarly, zinc application had no significant
effect on seed protein content, while salinity stress significantly decreased protein content. Decreasing protein percentage with increasing salinity could be attributed to the disturbance in nitrogen metabolism or to inhibition of nitrate absorption. It has been stated that the reduction in nitrogen under saline conditions might be due to the reduction of absorbed water and a decrease in root permeability (Strogonov et al., 1970). Medhat (2002), reported that salinity stress induces changes in the ion content of plant's cells, which in turn induce changes in the activity of certain metabolic systems that might have serious consequences for protein.

Mineral composition

Nitrogen content was not affected by zinc application while nitrogen fertilizer increased nitrogen content especially in the first year. This could be due to the fact that nitrogen availability increases nitrogen uptake. By contrast salinity diminished protein synthesis and accumulation (Tables 3 and 4). Higher accumulations of Na+ and Cl− concentrations result in decreased protein content (Sultana et al., 1999) and inhibit the growth of plants. Phosphorus content increased on account of nitrogen fertilizer just in the second year but it decreased due to salinity in both years (Tables 3 and 4). As for K, Ca and Mg, salinity significantly decreased concentration of these elements. Large increase in Na+, Cl− concentrations and Na/K ratio was observed due to salinity stress. However, despite large accumulation of Na+ and Cl− in plant tissues, effects of nitrogen and zinc on tissue concentrations of K, Ca, Mg, Na, Cl and Na/K ratio were not significant. Decline in root uptake efficiency of these nutrients with salt application may be attributed to internal osmotic adjustment of the seedlings in response to osmotic stress (Yang et al., 1990; Saneoka et al., 2001). These findings are in agreement with other reports suggesting that salt stress increases the Na/K ratio of green bean (Yasar et al., 2006), wheat (Hu et al., 2006) and legume (Amador et al., 2007). We can state that root cell membrane permeability decreased under salinity stress which might be related to the functions of Ca in cell membranes. From this point of view, external Na concentrations could demolish cell membrane and inhibit nutrition uptake or translocation.

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