Physiological and morphological responses of sugar beet (Beta vulgaris L.) subjected to nano-boron oxide at different growth stages

Alireza Pirzad*, Mahmood Mazlomi Manyandi, Razieh Khalilzadeh

1 Department of Plant Production and Genetics, Faculty of Agriculture and Natural Resources, Urmia University, Urmia, Iran

ABSTRACT The deficiency and toxicity of boron may lead to noticeable yield reduction and quality loss of sugar beet. To evaluate the effects of nano-boron on the root yield and quality of sugar beet, factorial experiment was conducted based on a randomized complete block design with three replications at Urmia University. Treatments were spraying of nano-boron oxide (0, 2, 3 and 4 g L\(^{-1}\)) at different growth stages (20, 40, 60, 80 and 100% of ground cover). Increasing levels of boron up to 3 g L\(^{-1}\) resulted in the highest SPAD (Soil Plant Analysis Development) value, leaf number and relative water content. With optimal leaf area (86.47 cm\(^2\)), the highest yields of root, sugar, white sugar, and technological sugar (144.53, 28.23, 26.19 and 25.32 t ha\(^{-1}\)) were determined in the treatment of 4 g L\(^{-1}\) boron at 40% of ground covered. Increasing nano-boron level under different growth stages increased sugar and white sugar contents, on contrary impurities (Na, K and α-amino-N) loss and molasses sugar percentage were decreased. More than 3 g L\(^{-1}\) nano-boron exhibited the highest values of purity. Application of 3 g L\(^{-1}\) boron at 40% of ground cover increased sugar and white sugar contents by 12.45 and 18.72%, respectively.

ARTICLE INFORMATION
Submitted 18 December 2019.
Accepted 03 February 2020.
*Corresponding author
E-mail: a.pirzad@urmia.ac.ir

*Corresponding author
E-mail: a.pirzad@urmia.ac.ir

ARTICLE
Physiological and morphological responses of sugar beet (Beta vulgaris L.) subjected to nano-boron oxide at different growth stages

Introduction

Sugar beet (Beta vulgaris L., Amaranthaceae) is one of the important sugar crops with high yield, high adaptability and high biological activity. It is also rich in minerals and organic nutrients (Grzegorzewski et al. 2017). Although sugar beet is well adapted to a wide range of growing conditions and soils, nutritional disorders caused by boron deficiency are quite common (Dordas 2007). Sustainable production requires the efficient use of inputs including adequate and balanced fertilization of both macro- and micro-nutrients (Singh et al. 2017). Deficiency of soil nutrients such as nitrogen (N), phosphorus (P), potassium (K), zinc (Zn) and boron (B) should be added to the rhizosphere according to plant needs and has been known as the major limitations in beet crop production (Abidow 2012). Boron is unique among the essential mineral micronutrients because of normal presence in soil solution as a non-ionized molecule over the vast pH range (Hirparan et al. 2018). It is also essential for plant in development and growth (Abido 2012) through the cell wall structure, membrane integrity and function (Nymora et al. 2019), sugar translocation from source to sink (Rawashdeh and Sala 2013) physiological functions such as carbohydrates metabolism, indole acetic acid metabolism, formation amino acid (Singh et al. 2017), nitrogen fixation (Rawashdeh and Sala 2013) and photosynthetic pigments (Abd El-hady 2017).

Deficiency of boron causes reduction in photosynthesis due to disturbs the activities of proton pumping ATPase and electron transport chain (Nadeem et al. 2019; Rehman et al. 2018), inhibition of leaf expansion, inhibitions root elongation through limiting mitosis and cell enlargement and division (Gemici et al. 2003), early enlargement due to clear limitation of its phloem mobility and reduced growth of new shoots and leaf (Ullah et al. 2013; Ali et al. 2015). Without an adequate supply and consumption of boron in large quantities, it may lead to marked yield reduction and quality loss of sugar production in soil application (Abbas et al. 2014; Tlili et al. 2017; Armin and Asghari-pour 2012). Because boron has been considered to have only limited phloem mobility and cannot readily be redistributed within the plant (Brown et al. 1999), the amount of yield losses directly depends upon the duration of deficiency and the plant growth stage at which it occurs (Ali et al. 2015). Deficiencies of boron result in many anatomical, biochemical and physiological changes...
in plants (Ali et al. 2015). Therefore, the management of boron in soil has become a worldwide agricultural problem in the recent years (Tlili et al. 2017).

Foliar fertilization has the advantage of low application rates, treated rapidly, uniform distribution and quick plant responses to applied nutrients (Asad et al. 2003; Saadati et al. 2013). Moreover, a number of previous studies have increased the significance of the role of foliar boron application in the productivity of crop plants (Perica et al. 2001; Dordas 2006; Abido 2012; Kristek et al. 2006). Also, to get the desired results, nanomaterial can be utilized by foliar application with much-decreased concentration (Prasad et al. 2012). After entering the cells the nanoparticles transport from one cell to another through plasmodesmata. The chemical and biological activities of most substances increase at the nanoscale (Dewdar et al. 2018). Root yield, sugar percentage significantly increased by increasing boron doses (Abbas et al. 2014). Dordas et al. (2006) reported that spraying of boron lead to a higher quality of sugar and root yield compared to the time using boric acid mixed with soil. El-Geddawy and Makhlouf (2015) found that there was a significant positive increase in root diameter and root length of sugar beet due to the gradual increase in the spraying concentration of boron from 105 to 210 ppm. Armin and Agharipour (2012) reported that the maximum root yield and sugar percentage was achieved by foliar application of 12% boric acid. Abd El-hady (2017) reported that root and sugar yields were increased by 19.4% and 39.5% compared with control treatment.

A better understanding of the physiological basis of the response of sugar beet may help in programs aiming to evaluate yield. Therefore, this work aimed to evaluate the effects of different amounts and time of nano-boron oxide spraying on the quantitative and qualitative aspects of sugar beet.

**Materials and Methods**

**Site and experimentation**

All experiments were conducted during 2011-2012 cropping seasons in North-Western Iran (i.e. Naghadeh (27°45' N latitude and 22°37' E longitude; Alt 1300 m) and were situated in the wet zone with moderate winter and hot summer. The experimental design was randomized factorial experiment based on a randomized complete block design with three replications. The soil type was silty loam and possessed around 7.95 pH, EC about 2.3 dS m⁻¹, total organic C = 1.20% and Zn = 32 mg kg⁻¹. The experimental soil was fertilized with 250 kg N ha⁻¹ in the form of urea (was applied as ½ at sowing, ½ at 6-8 leaf), 250 kg P ha⁻¹ in the form of triple superphosphate, and 100 kg K ha⁻¹ in the potassium nitrate at planting time. Experiment factors were the amount of nano-boron (Nano-B) oxide concentration (0, 2, 3 and 4 g L⁻¹ of nano chelate powder, with 99.5% of purity and 80 nm particle size, obtained from Khazra Company containing 9% chelated boron, absorbable at pH 3-11, and completely soluble in water) and spraying time included (20, 40, 60, 80 and 100 of ground cover by plant canopy). Each plot was consisted of 5 rows with 5 m long. The inter row and intra-row spacing was 10 and 15 cm, respectively. The sugar beet cultivar (Montarosa cv. a commonly grown cultivar of sugar beet in Naghade area) was sown at the depth of 20 cm on May 10.

**Measurements of quantity and quality parameters of sugar beet**

Harvesting was done manually 180 days after sowing (DAS). In order to measure the root length (cm), root diameter (cm) and leaf number traits, 10 plants in each plot were randomly harvested. Root yield (t ha⁻¹) was obtained from plants harvested of 5 m² in each experimental unit and juice quality characteristics were analyzed. The percentage of sucrose was determined according to Le-Docte (1927). Sodium and potassium (%) were determined by using a flame photometer (Model 410 Classic), nitrogen was determined according to the semi-micro Kjeldahl method (Model NA 1500) (Edwards 2014). Total soluble solids (TSS%) was measured in fresh roots using hand refractometer (model REF-113ATC). Juice purity% was also determined as a ratio between sucrose% and TSS% according to Carruthers and Oldfield (1961).

Sugar yield and root quality were calculated via the following equations:

\[
SY (\text{t ha}^{-1}) = RY (\text{t ha}^{-1}) \times SC(\%)
\]

Where, SY: sugar yield; RY: root yield; SC: sugar content.

\[
MS = 0.12 (K + Na) + 0.24(\alpha\text{-amino-N}) + 0.48
\]

Where, MS: molasses sugar (%); K: potassium (mmol/100 g root), Na: sodium (mmol/100 g root); \(\alpha\text{-amino-N}: \) alpha-amino-nitrogen (mmol/100 g root) (Buchholz et al. 1995).

\[
AC: \frac{(K + Na)}{N}.
\]

Where, AC: alkalinity coefficient.

\[
WSC(\%) = SC(\%) - MS(\%)
\]

Where, WSC: white sugar content.

\[
WSY (\text{t ha}^{-1}) = RY \times WSC(\%)
\]

Where, WSY: white sugar yield.
Where, WSY: white sugar yield.

\[
TSY (t \text{ ha}^{-1}) = RY (t \text{ ha}^{-1}) \times [SC (\%) - \text{loss of sugar productivity (\%)}]
\]

Where, TSY: technological sugar yield (Buchholz et al. 1995).

\[
LSP (\%) = MS (\%) + 0.6
\]

Where, LSP: loss of sugar productivity (Carruthers et al. 1961).

\[
\text{Sugar productivity (SP)} = \frac{SC - \text{loss of sugar productivity}}{SC}
\]

Where, SP: sugar productivity.

The leaf length was measured as the distance between the beginning of leaf formation on the leaf stem and the top of the leaf. The leaf width was measured at its widest point with a ruler. Based on measured leaf width (W, mm) and leaf length (L, mm) the area of each leaf (A, mm\(^2\)) is calculated using the following relationship (Mirschel 2018):

\[
A = W \times L \times 0.675
\]

The chlorophyll content of leaves was estimated with a SPAD-502 (Konica Minolta Sensing, Osaka, Japan) (Jifon et al. 2019). Relative water content was estimated according to the method of Tambussi et al. (2005).

Analysis of variance (ANOVA) and means comparison on data was performed using the SAS Statistical Package Program. Least Significant Difference (LSD) method was used to test the differences between means comparison of main effects and interactions.

**Results**

Analysis of variance showed that the significant interaction effect between Nano-B concentration and spraying stage on the relative water content (RWC), leaf area, root length, root and sugar yield, white and technological sugar yield, sugar content, white sugar content, sugar productivity, loss of sugar productivity, sodium, potassium, \(\alpha\)-amino-N and molasses sugar. There was a significant effect of Nano-B foliar application on the alkalinity coefficient (Table 1). SPAD, leaf number, root diameter, purity\% and TSS\% were affected by the Nano-B concentration and spraying stage (Table 1).

**Chlorophyll index, relative water content, leaf area and leaf number**

The maximum of SPAD (73.50) and leaf number (52.26) was obtained in B\(_3\) and B\(_4\), respectively (Table 1). Chlorophyll index was increased about by 34.71\% in 3 g L\(^{-1}\) Nano-B (Table 1). The highest content of SPAD and leaf number markedly increased from spraying of Nano-B at 80 of ground cover as G\(_4\) (Table 1). However, the difference in mention traits was not statistically significant in a comparison between G\(_2\), G\(_3\), and G\(_4\). The increasing of Nano-B at all growing stages up to 3 g L\(^{-1}\) resulted in the highest RWC. The highest and lowest RWC was respectively achieved in B\(_4\)G\(_4\) and B\(_3\)G\(_4\) (Table 2). A gradual increase in leaf area as growth stages improved up to G\(_4\) was recorded regardless of boron levels. The application of B\(_3\)G\(_4\) caused an increase in leaf area of 60\% in comparison with B\(_1\)G\(_4\).

**Root length and diameter**

It could be noticed that increasing boron rates significantly increased root diameter. The plant sprayed with 3 g L\(^{-1}\) of nano-boric acid revealed the highest root diameter (12.70 cm). Data also cleared that the late application of boron (G\(_4\)) recorded the highest value of root diameter (12.79 cm). Results showed that, the crops were fertilized early or late at different rates of boron, had any considerable differences on root length. Application of 2 g L\(^{-1}\) at 100\% of ground covered (B\(_2\)G\(_5\)), produced the highest root length (38.16 cm), while the lowest value (25.50 cm) was recorded in the B\(_1\)G\(_1\) (Table 2).

**Sugar beet yields**

Application of 4 g boron L\(^{-1}\) at the early stage of plant (40\% ground cover), can significantly increase root yield (144.53 t ha\(^{-1}\)), sugar yield (28.23 t ha\(^{-1}\)), white sugar yield (26.19 t ha\(^{-1}\)), and technological sugar yield (25.32 t ha\(^{-1}\)) to the highest amounts. Spraying nano-boric acid at the levels of B\(_3\), B\(_2\), and B\(_1\) increased root yield by about 29\%, 48\% and 79\% at early growth stage (G\(_2\)), as compared to control treatment (B\(_1\)), respectively. However, Abd El-hady (2017) reported that B element (1.0 kg B/ha) was increased root and sugar yields by 19.4\% and 39.5\% compared with control treatment.

**Quality of sugar beet**

The highest sugar and white sugar content were found to be in the B\(_3\)G\(_2\) treatment with average of 19.86\% and 18.39\%, respectively. However, their effects were also similar to B\(_4\)G\(_4\). Also, the lowest of SC and WSC are related to B\(_1\)G\(_1\) treatment with an average of 15.80\% and 14.14\%, respectively (Table 3). Compared to the control, spraying 3 g L\(^{-1}\) Nano-B at 40\% of ground covered improved SC and WSC by 12.45\% and 15.37\%, respectively. Data in Table 1
noticeably showed that $B_4$ and $B_3$ treatments recorded the highest values of purity percentage by 95.72% and 92.12%, respectively. It is worth mentioning that, considerable differences in purity% were not significant (Table 1). The later application at $G_5$ insignificantly surpassed the earlier application at $G_5$ in effecting purity%. Increasing the doses of B application from 0 to 4 g L$^{-1}$ provided the lowest LSP and MS 1.79 and 1.19 % with a decrease of 20.44% and 27.87%, respectively at the 60% of ground covered.

**Root impurities (K, Na and α-amino-N) and AC**

The values of impurities differed greatly due to the different treatments of time and boron rates (Table 3). There was a negative relationship between impurities and boron.

### Table 1. Effects of nano-boron oxide concentration and spraying time (growth stage) on growth and yield of sugar beet.

<table>
<thead>
<tr>
<th>Nano-B (B)</th>
<th>SPAD</th>
<th>RWC (%)</th>
<th>Leaf number</th>
<th>Leaf area (mm$^2$)</th>
<th>Root length (cm)</th>
<th>Root diameter (cm)</th>
<th>Root yield</th>
<th>Sugar yield</th>
<th>WSY</th>
<th>TSY</th>
</tr>
</thead>
<tbody>
<tr>
<td>$B_1$ = no nano-B as control</td>
<td>54.56b</td>
<td>73.98b</td>
<td>33.73c</td>
<td>49.52c</td>
<td>26.70c</td>
<td>10.73b</td>
<td>88.77d</td>
<td>14.93d</td>
<td>13.39d</td>
<td>12.86d</td>
</tr>
<tr>
<td>$B_2$ = 2 g L$^{-1}$</td>
<td>59.55b</td>
<td>82.07a</td>
<td>41.46b</td>
<td>67.63b</td>
<td>30.53b</td>
<td>11.23b</td>
<td>108.25c</td>
<td>18.92c</td>
<td>17.26c</td>
<td>16.61c</td>
</tr>
<tr>
<td>$B_3$ = 3 g L$^{-1}$</td>
<td>73.50a</td>
<td>84.82a</td>
<td>52.26a</td>
<td>74.55ab</td>
<td>31.00b</td>
<td>12.70a</td>
<td>127.65a</td>
<td>24.15a</td>
<td>22.42a</td>
<td>21.65a</td>
</tr>
<tr>
<td>$B_4$ = 4 g L$^{-1}$</td>
<td>68.35a</td>
<td>83.27a</td>
<td>49.33a</td>
<td>82.85a</td>
<td>34.68a</td>
<td>12.70a</td>
<td>127.65a</td>
<td>24.15a</td>
<td>22.42a</td>
<td>21.65a</td>
</tr>
</tbody>
</table>

LSD (p<0.05) 5.20 4.08 4.96 10.47 2.49 1.24 8.05 1.70 1.60 1.55

**Growth stage (G)**

<table>
<thead>
<tr>
<th>G</th>
<th>SC (%)</th>
<th>WSC (%)</th>
<th>Purity %</th>
<th>SP</th>
<th>LSP</th>
<th>Na</th>
<th>K</th>
<th>α-amino-N</th>
<th>AC</th>
<th>MS</th>
<th>TSS %</th>
</tr>
</thead>
<tbody>
<tr>
<td>$G_1$ = 20% of ground cover</td>
<td>16.84b</td>
<td>15.11c</td>
<td>75.92c</td>
<td>0.861d</td>
<td>2.33a</td>
<td>1.20a</td>
<td>6.84a</td>
<td>1.18a</td>
<td>6.99c</td>
<td>1.73a</td>
<td>22.20a</td>
</tr>
<tr>
<td>$G_2$ = 40% of ground cover</td>
<td>17.50b</td>
<td>15.96b</td>
<td>82.83b</td>
<td>0.877c</td>
<td>0.79b</td>
<td>2.13b</td>
<td>0.85b</td>
<td>8.46b</td>
<td>1.53b</td>
<td>21.15b</td>
<td></td>
</tr>
<tr>
<td>$G_3$ = 60% of ground cover</td>
<td>18.54a</td>
<td>17.11a</td>
<td>92.12a</td>
<td>0.890b</td>
<td>2.03c</td>
<td>0.79b</td>
<td>5.77c</td>
<td>0.74b</td>
<td>8.97b</td>
<td>1.43c</td>
<td>20.18bc</td>
</tr>
<tr>
<td>$G_4$ = 80% of ground cover</td>
<td>18.90a</td>
<td>17.54a</td>
<td>95.72a</td>
<td>0.896a</td>
<td>1.96d</td>
<td>0.74b</td>
<td>5.61c</td>
<td>0.74c</td>
<td>8.73a</td>
<td>1.51b</td>
<td>21.54a</td>
</tr>
<tr>
<td>$G_5$ = 100% of ground cover</td>
<td>18.01</td>
<td>16.41ab</td>
<td>89.38a</td>
<td>0.877b</td>
<td>2.20a</td>
<td>0.52c</td>
<td>6.63a</td>
<td>0.93a</td>
<td>8.70a</td>
<td>1.60a</td>
<td>20.30bc</td>
</tr>
</tbody>
</table>

LSD (p<0.05) 0.69 0.67 4.90 0.005 0.07 0.117 0.45 0.118 1.12 0.07 0.96

### Table 1 (Continued). Effects of nano-boron oxide concentration and spraying time (growth stage) on growth and yield of sugar beet.

| B × G | ns | ** | ns | * | ** | ns | * | ** | ns | * | ** |
| C.V. | 10.14 | 5.18 | 14.51 | 14.96 | 8.28 | 11.89 | 7.12 | 8.31 | 8.55 | 8.65 |

ns: show no significant differences.
* and **: show significant differences at 0.05 and 0.01 probability level, respectively.

SC: sugar content; WSC: white sugar content; K: potassium; Na: sodium; α-amino-N: alpha-amino-nitrogen; AC: alkalinity coefficient; TSS: total soluble solids; SP: sugar productivity; LSP: loss of sugar productivity. MS: molasses sugar.
foliar application. Data in Table 3 exposed that, increasing the boron concentrations from 0 to 4 g L\(^{-1}\) contributed the last potassium and \(\alpha\)-amino-N content of 4.58 and 0.45 mmol/100 g root at 60% growth stage. On the other hand, without the application of boron (B1) resulted in the maximum mean values of K (7.69) and \(\alpha\)-amino-N (1.47) content at B1G5. The significant lowest sodium content 0.50 and 0.49 mmol/100 g root related to a reduction in impurity 60.37% and 66.66%, at the application of B4G3 and B4G4, respectively.

Discussion

Foliar application of Nano-B at 3 g L\(^{-1}\) resulted in a consistent improvement in vegetative growth of sugar beet, but on increasing Nano-B concentration up to B4, vegetative growth decreased compared with B3. In addition, the increases of yield-related responses like chlorophylls and leaf area (Table 2) of sugar beet at high Nano-B concentration (B3 and B4) could be reflected upon the increase of sugars percentage and reduction of impurities (Table 3), so, the optimal leaf area value for root and sugar yields was 86.47 cm\(^2\) at B4G4. Leaf number and low chlorophyll content at high boron concentration are associated with toxicity of this element (Armin and Asgharipour 2012). Adequate boron supply through foliar application improved the chlorophyll content, leaf number and leaf area enabling them to capture more light and produce more assimilate for loading to root. Ullah et al. (2013) reported that B deficiency causes to reduced growth of new leaf and shoot due to clear limitation of boric acid phloem mobility. Such enhancement effect of B could be related to the favorable influence of them on photosynthetic pigments (Wanas 2002; Abd El-hady et al. 2017), metabolism, enzyme activity (El-Sherbeny et al. 2007), photosynthesis efficiency (Abou El-Yazied and Mady 2012) which in turn encourage vegetative growth and increasing dry matter production. Also, this enhancement could be an indicator of expectable high sugar beet yield. Also, Abd El-hady et al. (2017) informed that these results might be attributed to that B is an essential element for photosynthetic pigments, where it increases CO\(_2\) fixation, rates of photosynthetic \(\text{O}_2\) evolution and decreases respiration and the activities of oxidative pentose phosphate enzymes.

It seems that the increase in root diameter at 3 g L\(^{-1}\) of

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Growth stage</th>
<th>RWC (%)</th>
<th>Leaf area (mm(^2))</th>
<th>Root length (cm)</th>
<th>Root yield</th>
<th>Sugar yield</th>
<th>WSY (t ha(^{-1}))</th>
<th>TSY (t ha(^{-1}))</th>
</tr>
</thead>
<tbody>
<tr>
<td>B1</td>
<td>G1</td>
<td>77.34±3.65</td>
<td>52.10±6.53</td>
<td>25.50±2.50</td>
<td>82.87±1.17</td>
<td>13.08±1.05</td>
<td>11.17±0.89</td>
<td>11.21±0.85</td>
</tr>
<tr>
<td></td>
<td>G2</td>
<td>79.18±2.06</td>
<td>50.66±2.51</td>
<td>26.66±1.52</td>
<td>80.93±1.88</td>
<td>14.28±0.42</td>
<td>12.90±0.39</td>
<td>12.41±0.39</td>
</tr>
<tr>
<td></td>
<td>G3</td>
<td>75.65±2.64</td>
<td>58.61±6.88</td>
<td>25.33±1.52</td>
<td>90.56±9.19</td>
<td>15.06±1.72</td>
<td>13.55±1.54</td>
<td>13.01±1.49</td>
</tr>
<tr>
<td></td>
<td>G4</td>
<td>79.16±3.50</td>
<td>57.57±2.97</td>
<td>27.50±2.17</td>
<td>100.88±8.64</td>
<td>16.39±1.17</td>
<td>14.68±1.09</td>
<td>14.08±1.04</td>
</tr>
<tr>
<td></td>
<td>G5</td>
<td>58.57±5.09</td>
<td>28.69±1.69</td>
<td>36.16±1.89</td>
<td>88.61±8.10</td>
<td>15.84±1.66</td>
<td>14.13±1.47</td>
<td>13.60±1.42</td>
</tr>
<tr>
<td>B2</td>
<td>G1</td>
<td>81.64±3.22</td>
<td>51.49±6.04</td>
<td>31.33±1.25</td>
<td>98.36±6.93</td>
<td>17.43±1.02</td>
<td>15.99±0.96</td>
<td>15.40±0.92</td>
</tr>
<tr>
<td></td>
<td>G2</td>
<td>82.22±3.87</td>
<td>66.32±10.35</td>
<td>27.50±2.00</td>
<td>112.21±5.77</td>
<td>19.13±0.73</td>
<td>17.46±0.67</td>
<td>16.79±0.65</td>
</tr>
<tr>
<td></td>
<td>G3</td>
<td>82.33±1.86</td>
<td>74.23±15</td>
<td>34.56±3.84</td>
<td>106.85±10.80</td>
<td>19.51±1.46</td>
<td>17.91±1.36</td>
<td>17.26±1.30</td>
</tr>
<tr>
<td></td>
<td>G4</td>
<td>81.25±1.16</td>
<td>84.36±5.99</td>
<td>34.83±2.84</td>
<td>115.25±10.36</td>
<td>19.68±2.05</td>
<td>17.87±1.99</td>
<td>17.18±1.93</td>
</tr>
<tr>
<td></td>
<td>G5</td>
<td>82.93±2.44</td>
<td>61.78±17.16</td>
<td>38.16±2.25</td>
<td>106.8±7.77</td>
<td>18.87±2.30</td>
<td>17.09±2.10</td>
<td>16.44±2.05</td>
</tr>
<tr>
<td>B3</td>
<td>G1</td>
<td>85.23±4.76</td>
<td>53.45±4.49</td>
<td>29.16±1.52</td>
<td>111.17±3.47</td>
<td>19.11±0.85</td>
<td>17.58±0.78</td>
<td>16.92±0.78</td>
</tr>
<tr>
<td></td>
<td>G2</td>
<td>88.86±9.10</td>
<td>74.83±9.15</td>
<td>25.66±2.36</td>
<td>119.89±10.14</td>
<td>23.81±2.23</td>
<td>22.05±2.10</td>
<td>21.34±2.05</td>
</tr>
<tr>
<td></td>
<td>G3</td>
<td>85.41±1.38</td>
<td>76.06±12.47</td>
<td>33.00±4.35</td>
<td>119.06±13.26</td>
<td>22.97±2.30</td>
<td>21.82±2.19</td>
<td>20.57±2.13</td>
</tr>
<tr>
<td></td>
<td>G4</td>
<td>83.56±10.56</td>
<td>92.13±18.71</td>
<td>32.16±2.75</td>
<td>118.42±5.98</td>
<td>21.37±0.59</td>
<td>19.59±0.46</td>
<td>18.88±0.43</td>
</tr>
<tr>
<td></td>
<td>G5</td>
<td>83.51±2.20</td>
<td>79.30±15.30</td>
<td>30.83±3.84</td>
<td>116.90±6.79</td>
<td>21.38±1.89</td>
<td>19.74±1.77</td>
<td>19.04±1.73</td>
</tr>
<tr>
<td>B4</td>
<td>G1</td>
<td>82.78±2.47</td>
<td>65.57±9.14</td>
<td>29.50±1.32</td>
<td>103.38±7.20</td>
<td>19.49±1.16</td>
<td>18.02±1.10</td>
<td>17.40±1.07</td>
</tr>
<tr>
<td></td>
<td>G2</td>
<td>83.66±0.31</td>
<td>86.47±10.87</td>
<td>31.83±1.75</td>
<td>144.53±9.55</td>
<td>28.23±0.26</td>
<td>26.19±2.01</td>
<td>25.32±1.98</td>
</tr>
<tr>
<td></td>
<td>G3</td>
<td>82.99±0.46</td>
<td>86.29±8.96</td>
<td>28.1±6.11</td>
<td>135.54±6.01</td>
<td>25.24±2.89</td>
<td>23.62±2.85</td>
<td>22.80±2.82</td>
</tr>
<tr>
<td></td>
<td>G4</td>
<td>82.74±1.99</td>
<td>89.36±8.50</td>
<td>30.33±2.75</td>
<td>127.54±2.52</td>
<td>24.14±0.60</td>
<td>22.49±0.56</td>
<td>21.72±0.54</td>
</tr>
<tr>
<td></td>
<td>G5</td>
<td>81.74±2.03</td>
<td>86.56±14.19</td>
<td>36.33±4.01</td>
<td>127.28±7.60</td>
<td>23.67±1.56</td>
<td>21.81±1.44</td>
<td>21.04±1.40</td>
</tr>
</tbody>
</table>

LSD\(_{0.05}\) 6.94 16.97 4.21 13.01 2.73 2.58 2.52

B\(_2\) indicates no application; B\(_3\), B\(_4\) and B\(_5\) indicate application of 2, 3 and 4 g L\(^{-1}\) of Nano-B, respectively. G\(_1\), G\(_2\), G\(_3\), G\(_4\) and G\(_5\) indicate foliar application Nano-B at 20, 40, 60, 80 and 100% of ground cover, respectively.

Table 2. Means comparison the effects of Nano-B concentration and spraying time (growth stage) on RWC, leaf area, root length, root yield, sugar yield, WSY and TSY of sugar beet.
Nano-B by means of high leaf number and chlorophyll content and efficient assimilates portioning towards sink parts. These results could be explained by the role of boron in plant metabolism, development and growth (Rawashdeh and Sala 2013; Abido 2012), cell wall formation and meristematic tissue extension and cell elongation the root (Nalini et al. 2013). Cell enlargement and increase in a number of cells contribute to the increase of yield can be due to the role of boron in the biosynthesis of auxin in the meristematic activity and increase in the IAA-oxidase activity. Similar observations were recorded by Abdelaal et al. (2015) and Dugger (1973) in sugar beet.

Armin and Asgharipour (2012) stated that the maximum root yield and sugar percentage was achieved by foliar application of 12% boric acid. Considering this, Nano-B spraying may be used to enhance root and sugar yield, resulting in reduce boron fertilizer. Foliar Nano-B application predominantly affects at vegetative growth compared with reproductive growth in sugar beet. Results in Table 2 exhibited that sugar yield and white sugar yield was significantly improved by increasing of B from B1 to B5. These results were true in the five growth stages. While that sugar content was decreased by B5 when compared with B1 treated plants (Table 2), the increase in sugar yield accompanying high boron level might have been due to the increase in root yield as well as sucrose content. These results are in agreement by those of Gezgin et al. (2001).

It seems that, better translocation of photosynthates from high leaf area (Table 2) and higher dry matter accumulation with high root length led to increasing in root yield (Table 2). The increase in tops and roots fresh and dry weights, caused by boron application, could be attributed to the stimulating effect of boron on the photosynthesis process in the plants such as translocation of sugar and carbohydrates of assimilates from the top to root, which leads to increase in root and sugar yield. On the other hand, when photosynthetic activity is high, any factor that increases the leaf area may have a positive effect on WSY. The enhancement of dry matter in sugar beet roots may be attributed to the improvement of leaf area, leaf number, RWC and chlorophyll content which results in improvement of growth-related traits such as root length and root diameter, and consequently root yield sugar yield (Table 2). Similar results were also observed by Abdel-Motagally (2015) and Mohammadian et al. (2014) who reported that early beginning of photosynthetic

<table>
<thead>
<tr>
<th>Treatment</th>
<th>SC (%)</th>
<th>WSC (%)</th>
<th>SP</th>
<th>LSP</th>
<th>Na</th>
<th>K</th>
<th>N</th>
<th>MS</th>
<th>(mmol/100g root)</th>
</tr>
</thead>
<tbody>
<tr>
<td>B1</td>
<td>G1</td>
<td>15.80±0.20</td>
<td>14.14±0.27</td>
<td>0.857±0.006</td>
<td>2.25±0.085</td>
<td>0.94±0.16</td>
<td>6.93±0.71</td>
<td>0.96±0.11</td>
<td>1.65±0.081</td>
</tr>
<tr>
<td></td>
<td>G2</td>
<td>17.66±0.83</td>
<td>15.94±0.74</td>
<td>0.868±0.0025</td>
<td>2.32±0.094</td>
<td>0.96±0.27</td>
<td>7.03±0.87</td>
<td>1.17±0.24</td>
<td>1.72±0.096</td>
</tr>
<tr>
<td></td>
<td>G3</td>
<td>16.63±0.73</td>
<td>14.97±0.75</td>
<td>0.863±0.0077</td>
<td>2.25±0.058</td>
<td>1.26±0.22</td>
<td>6.33±0.59</td>
<td>1.11±0.19</td>
<td>1.65±0.060</td>
</tr>
<tr>
<td></td>
<td>G4</td>
<td>16.26±0.41</td>
<td>14.56±0.37</td>
<td>0.859±0.003</td>
<td>2.29±0.070</td>
<td>1.47±0.101</td>
<td>6.23±0.24</td>
<td>1.22±0.14</td>
<td>1.69±0.067</td>
</tr>
<tr>
<td></td>
<td>G5</td>
<td>17.86±0.64</td>
<td>15.94±0.58</td>
<td>0.859±0.002</td>
<td>2.52±0.070</td>
<td>1.37±0.061</td>
<td>7.69±0.79</td>
<td>1.47±0.14</td>
<td>1.92±0.068</td>
</tr>
<tr>
<td>B2</td>
<td>G1</td>
<td>17.73±0.30</td>
<td>16.26±0.23</td>
<td>0.883±0.002</td>
<td>2.07±0.072</td>
<td>0.66±0.047</td>
<td>5.84±0.28</td>
<td>0.85±0.21</td>
<td>1.46±0.071</td>
</tr>
<tr>
<td></td>
<td>G2</td>
<td>17.06±0.65</td>
<td>15.8±0.65</td>
<td>0.877±0.0049</td>
<td>2.08±0.011</td>
<td>0.76±0.035</td>
<td>5.93±0.13</td>
<td>0.84±0.03</td>
<td>1.48±0.011</td>
</tr>
<tr>
<td></td>
<td>G3</td>
<td>18.30±0.79</td>
<td>16.79±0.77</td>
<td>0.885±0.0045</td>
<td>2.10±0.043</td>
<td>0.83±0.100</td>
<td>5.96±0.19</td>
<td>0.86±0.19</td>
<td>1.50±0.045</td>
</tr>
<tr>
<td></td>
<td>G4</td>
<td>17.06±0.50</td>
<td>15.49±0.54</td>
<td>0.872±0.0073</td>
<td>2.17±0.081</td>
<td>0.81±0.081</td>
<td>9.56±0.32</td>
<td>0.86±0.14</td>
<td>1.57±0.081</td>
</tr>
<tr>
<td></td>
<td>G5</td>
<td>17.33±0.90</td>
<td>15.69±0.83</td>
<td>0.871±0.034</td>
<td>2.23±0.075</td>
<td>0.91±0.085</td>
<td>7.05±0.52</td>
<td>0.83±0.05</td>
<td>1.63±0.072</td>
</tr>
<tr>
<td>B3</td>
<td>G1</td>
<td>17.20±0.80</td>
<td>15.82±0.78</td>
<td>0.885±0.0049</td>
<td>1.97±0.053</td>
<td>0.63±0.062</td>
<td>5.59±0.30</td>
<td>0.60±0.09</td>
<td>1.37±0.057</td>
</tr>
<tr>
<td></td>
<td>G2</td>
<td>19.86±0.80</td>
<td>18.39±0.79</td>
<td>0.896±0.0040</td>
<td>2.06±0.025</td>
<td>0.57±0.028</td>
<td>6.30±0.24</td>
<td>0.67±0.07</td>
<td>1.46±0.021</td>
</tr>
<tr>
<td></td>
<td>G3</td>
<td>19.33±0.98</td>
<td>17.90±0.98</td>
<td>0.894±0.0061</td>
<td>2.02±0.050</td>
<td>0.69±0.060</td>
<td>5.56±0.27</td>
<td>0.80±0.20</td>
<td>1.42±0.052</td>
</tr>
<tr>
<td></td>
<td>G4</td>
<td>18.06±0.50</td>
<td>16.56±0.50</td>
<td>0.883±0.0040</td>
<td>2.10±0.052</td>
<td>0.83±0.015</td>
<td>5.99±0.36</td>
<td>0.84±0.09</td>
<td>1.50±0.052</td>
</tr>
<tr>
<td></td>
<td>G5</td>
<td>18.26±0.57</td>
<td>16.86±0.55</td>
<td>0.890±0.0023</td>
<td>2.00±0.20</td>
<td>0.67±0.105</td>
<td>5.41±0.21</td>
<td>0.78±0.12</td>
<td>1.39±0.019</td>
</tr>
<tr>
<td>B4</td>
<td>G1</td>
<td>18.86±0.64</td>
<td>17.44±0.66</td>
<td>0.893±0.0050</td>
<td>2.02±0.034</td>
<td>0.53±0.041</td>
<td>5.92±0.11</td>
<td>0.69±0.10</td>
<td>1.42±0.035</td>
</tr>
<tr>
<td></td>
<td>G2</td>
<td>19.53±0.90</td>
<td>18.11±0.91</td>
<td>0.896±0.0050</td>
<td>2.02±0.052</td>
<td>0.52±0.058</td>
<td>5.94±0.48</td>
<td>0.68±0.03</td>
<td>1.41±0.054</td>
</tr>
<tr>
<td></td>
<td>G3</td>
<td>18.60±1.56</td>
<td>17.40±1.58</td>
<td>0.903±0.0091</td>
<td>1.79±0.046</td>
<td>0.50±0.073</td>
<td>4.58±0.33</td>
<td>0.45±0.08</td>
<td>1.19±0.047</td>
</tr>
<tr>
<td></td>
<td>G4</td>
<td>18.93±0.23</td>
<td>17.63±0.25</td>
<td>0.899±0.0030</td>
<td>1.90±0.043</td>
<td>0.49±0.072</td>
<td>5.23±0.20</td>
<td>0.55±0.18</td>
<td>1.30±0.043</td>
</tr>
<tr>
<td></td>
<td>G5</td>
<td>18.60±0.52</td>
<td>17.13±0.55</td>
<td>0.888±0.0047</td>
<td>2.06±0.041</td>
<td>0.54±0.032</td>
<td>6.37±0.30</td>
<td>0.64±0.15</td>
<td>1.46±0.039</td>
</tr>
<tr>
<td>LSD0.05</td>
<td>1.207</td>
<td>1.20</td>
<td>0.0085</td>
<td>0.092</td>
<td>0.188</td>
<td>0.66</td>
<td>0.22</td>
<td>0.091</td>
<td></td>
</tr>
</tbody>
</table>

B1 indicates no application; B2, B3 and B4 indicate application of 2, 3 and 4 g L⁻¹ of Nano-B, respectively. G1, G2, G3, G4 and G5 indicate foliar application Nano-B at 20, 40, 60, 80 and 100% of ground cover, respectively.
transmission from leaf to root and consequently, it would increase WSY at the harvest time.

The rise of WSC may be due to the increase in sugar percentage and the reduction of impurities in terms of sodium, potassium and α-amino-N content (Table 3). Further, if optimum B is available at the early growth stage, the plant continues to partition it in the sink part as have been observed in this study. This finding suggested that sugar content is the key factor conferring B phloem mobility due to the bonding of sugar and boric acid (Liakopoulos et al. 2005). The Nano-boron facilitates the transport of sugars in the plants because it had a crucial role in the biosynthesis of auxin (Dugger 1973; Ullah et al. 2013). The high amount of juice purity would be desirable to provide sugar content. The important role of the boron element on the percentage of purity comes through its beneficial effect on the values of sucrose content (Table 3). Boron dominates in the early-stage, building up the highest TSS for the sugar beets (Table 1).

The result showed that, at low concentration of boron, a lower percentage of sugar had been achieved. Therefore, sugar in the form of molasses in these plants is higher in amount than that which has received enough boron (Table 3). Reduction of impurities like sodium, potassium and α-amino-N content in beet roots cause to decrease of sucrose molasses under application of a higher rate of boron application. These results are an agreement with the finging of Abbas et al. (2014) who reported that increasing the concentration of B cause to the reduction of sugar molasses. Hellal et al. (2009) showed that juice purity, sugar and root yield of sugar beet improved by increasing of B spraying which could be related to the reduction of sodium and potassium uptake in root juice.

The reasons for the improvement in sugar beet quality could be due to the fact that B plays a role in cell division, enhanced enzymatic activity, the membrane integrity, calcium uptake and carbohydrate metabolism (Nalini et al. 2013; Rawashdeh and Sala 2013). An increased boron supply decreases the nitrate levels via inhibiting transcript level in the roots and altering the nitrate transporter activity, leading to reduced plasma membrane enzymes activities (Camacho-Cristobal and Gonzalez-Fontes 2007). Evidence proposes that sugar alcohols synthesis and the later transport of the B-sugar alcohol compound in the phloem to sink tissues is the main factor that confers phloem B mobility to a plant species (Brown et al. 1999). The least accumulation of sodium at the later application (G4 and G6) could be due to the increased of leaf area at this time and facilitate the improvement of B absorption. Similar results were obtained by Abbas et al. (2014) who reported that spraying dates lead to significantly different in sodium content. Also, Armin and Asgharipour (2012) and Abbas et al. (2014) showed that boron application improved juice quality by declining K and Na content.

**Conclusion**

Application of Nano-B rates showed a significant increase in quantitative and qualitative sugar beet traits under study. The highest SPAD and RWC, leaf area and leaf number were observed in 3 g L⁻¹ of Nano-B resulted in a consistent improvement in vegetative growth of sugar beet but with increasing concentration of Nano-B mentioned parameters were decreased. The increasing of boron fertilizer at all growing stages resulting in the highest root yield, sugar content and white sugar content thus led to increasing sugar yield, white and technological sugar yield. The decrease in sucrose molasses in a high level of boron accompanying due to the reduction in impurities in terms of sodium, potassium and α-amino-N content in sugar beet roots. Therefore, B₄G₂ treatment (4 g L⁻¹ boron at 40% of ground covered) with the highest root and sugar yield may be recommended for the cultivation of sugar beet in terms of time and fertilizer saving.

**Acknowledgements**

We thank “Iran’s National Elites Foundation” for their help during the course of experimentation.

**References**


Nadeem F, Farooq M, Nawaz A, Ahmad R (2019) Boron improves productivity and profit ability of bread wheat...