**Project Title:** Evaluation of Soybean Tolerance to Salinity, Alkalinity, and the Combined Saline-alkalinity

**Objectives:**

* Developing methods used for screening soybean tolerance to salinity, alkalinity, and saline-alkalinity
* Evaluating tolerance of 50 soybean genotypes to the aforementioned three stresses
1. **Developing methods used for screening soybean tolerance to salinity, alkalinity, and saline-alkalinity**

‘Barnes’ and ‘A11’ were used in this objective. ‘Barnes’ and ‘A11’ seeds were germinated in a growth chamber for three days and then transformed to a hydroponic system with full strength Hoagland solution in a greenhouse. After a 7-day acclimation to the hydroponic system, soybean seedlings were exposed to salinity (Experiment 1), alkalinity (Experiment 2), or the combined saline-alkalinity conditions. (Experiment 3).

***Experiment 1 - Salinity.*** Salinity conditions were induced by amending NaCl or a Na2SO4:MgSO4 mixture (1:1, molar ratio) (0 – 16 dS m-1) to the Hoagland solution. Plants were grown in the saline solutions for three weeks. The experimental design was a split-plot design, with the combination of salt type x salt concentration being the whole-plot treatment arranged in a randomized complete block design with three replicates, and cultivar being the sub-plot treatment. Data were collected on shoot and root dry weight, root length, absolute water content (shoot fresh weight – dry weight, an indicator of leaf size), and relative shoot and root water content ((fresh weight – dry weight)/fresh weight x 100).

Root length decreased from 57.0 cm at 0 dS m-1 to 22.2 cm at 16 dS m-1. Plants under the SO42- treatment had a longer root, 40.1 cm, compared to those under the Cl- treatment (36.1 cm). No cultivar differences were observed in root length, averaged 38.1 cm. A salt type x salt concentration interaction was observed in shoot dry weight, root dry weight, absolute water content, and shoot water content. Plants under the NaCl treatments were severely damaged at 4 dS m-1 as shown in absolute water content (Figure 1) and mortality was observed at 8 – 16 dS m-1. In contrast, growth inhibition in the SO42- treated plants did not occur until 8 dS m-1. Plants under the SO42- treatment showed a higher shoot dry weight than those under the Cl- treatment at all salinity levels, except 0 dS m-1. Similar results were observed in shoot and root dry weight and relative shoot water content. Cultivar differences were observed in shoot dry weight and leaf size, in which ‘Barnes’ outperformed than ‘A11’ when data were pooled across salt type and concentration. Root water content was not influenced by any main factors nor their interaction. Eight dS m-1 was selected to screening for sulfate-salinity tolerance as this salinity level caused moderate plant damage.

Figure 1. Absolute water content of soybean seedlings grown under NaCl or Na2SO4:MgSO4 mixture (SO) (1:1, molar ratio) salinity. Uppercase letter indicates significant differences (*P* ≤ 0.05) between salt concentrations within the same salt type. Lowercase letter indicates significant differences (*P* ≤ 0.05) between the two salt types at the same salt concentration.

Plants under the NaCl treatment (0 and 4 dS m-1) and sulfate treatment (0 – 12 dS m-1) produced enough biomass for shoot nutrient analysis (K, Na, Ca, Fe, Mn, and Zn). No cultivar and salinity differences were observed in NaCl-treated plants in any nutrient element (data not shown). In contrast, Barnes’ had a lower level of Fe, Mn, and Zn, but higher Na than ‘A11’ under the sulfate-salinity (Table 1). Salinity effect was detected in all nutrients, except Fe and Mn.

Table 1. Shoot nutrient contents (mg/100 g dry weight) of soybean seedlings grown in a hydroponic system amended with a Na2SO4:MgSO4 mixture (1:1, molar ratio). Data were pooled across the salinity or cultivar.

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
|  | K |  Na | Ca | Fe | Mn |  Zn |
| Cultivar  |  |  |  |  |  |  |
| A11 | 2115.2az | 276.0b |  584.7a | 23.2a |  11.7a |  3.7a |
| Barnes | 2232.3a | 421.7a |  546.2a | 11.0b |  5.1b |  2.7b |
| Salinity (dS m-1) |  |  |  |  |  |  |
| 0 | 2372.8a |  44.7b | 1040.7a | 17.4a |  9.1a |  1.8b |
| 4 | 2400.4a | 234.3a |  605.5b | 14.9a |  7.2a |  3.9a |
| 8 | 2053.2b | 146.7ab |  499.6b | 15.5a |  9.4a |  4.3a |
| 12 | 2015.9b | 139.2ab |  337.5c | 21.9a |  8.3a |  2.5b |

zMeans followed by the same letter within the same main factor in a nutrient are not significantly different at *P* ≤ 0.05.

***Experiment 2 - Alkalinity.*** Alkalinity was induced by NaHCO3:Na2CO3 mixtures (5 mM) at 10:0, 8:2, 6:4, 4:6, 2:8, and 0:10 (molar ratio) ratio. A control treatment (i.e. without NaHCO3 and Na2CO3) was also included. pH of the treatments ranged from 6.5 (the control) to 9.4 (0:10, M:M). Plants were grown under the alkali conditions for four weeks. The experimental design was a split-plot design, with the alkali solution being the whole-plot treatment arranged in a randomized complete block design with three replicates, and cultivar being the sub-plot treatment.

Shoot dry weight decreased with increasing pH (Table 2). Similar results were observed in root biomass, root length, absolute water content, and shoot water content. Although biomass started to reduce at the 10:0 (molar ratio) treatment, the visual leaf damage (i.e. chlorosis) was more pronounced at the 2:8 and 0:10 (molar ratio) treatments as seen in the picture below (‘Barnes’ – left and ‘A11’ – right); thus, the 2:8 (molar ratio) treatment was selected to screen for alkalinity tolerance.

|  |  |
| --- | --- |
| C:\NDSU\Potential research projects\Soybean salinity and alkalinity\PICTURES\Two cultivars pH Run 1\IMG_4992.JPG | C:\NDSU\Potential research projects\Soybean salinity and alkalinity\PICTURES\Two cultivars pH Run 1\IMG_4988.JPG |

Table 2. Shoot dry weight of soybean seedlings grown under the alkali condition (NaHCO3:Na2CO3). No NaHCO3 and Na2CO3 were added in the control treatment. Data were pooled across cultivar.

|  |  |
| --- | --- |
|  | NaHCO3:Na2CO3 (molar ratio) |
|  | Control | 10:0 | 8:2 | 6:4 | 4:6 | 2:8 | 0:10 |
| Shoot dry weight (g) | 2.109az | 0.814bc  | 0.906b  | 0.768b-d  | 0.560b-d  | 0.487cd  | 0.414d |

zMeans followed by the same letter are not significantly different at *P* ≤ 0.05.

Shoot nutrient content was analyzed in all the treatments, except the 0:10 treatment due to low biomass. The general trends of nutrient contents under the alkaline condition were similar to that under the saline condition, in which ‘A11’ had higher nutrient content than ‘Barnes’ (except Na and Zn) and nutrient levels decreased with increasing alkalinity (except Na) (Table 3).

Table 3. Shoot nutrient contents (mg/100 g dry weight) of soybean seedlings grown under alkalinity in a hydroponic system. Data were pooled across alkalinity or cultivar.

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
|  | K | Na | Ca | Fe | Mn | Zn |
| Cultivar (C) |  |  |  |  |  |  |
| A11 | 2136.3a |  76.8a | 1077.3a |  10.3a |  7.5a | 0.9a |
| Barnes | 1898.6b |  72.6a |  960.7b | 7.1b |  5.1b | 0.9a |
| NaHCO3:Na2CO3 (molar ratio) |
| Control (no salt) | 3109.7a |  27.2d | 1376.7a | 22.4a |  10.8a | 2.7a |
| 10:0 | 1645.8c |  28.6d | 1007.2bc | 9.2b |  8.2b | 0.4c |
| 8:2 | 1730.1c |  40.2cd | 1057.8b |  7.6bc |  6.7bc | 0.4c |
| 6:4 | 1618.5c |  47.6c |  894.9d | 4.4c |  4.9cd | 0.3c |
| 4:6 | 1730.6c |  117.3b |  858.0d | 4.3c |  3.5d | 0.6c |
| 2:8 | 2270.1b |  187.1a |  919.4cd | 4.2c |  4.0cd | 1.0b |

zMeans followed by the same letter within the same main factor in a nutrient are not significantly different at *P* ≤ 0.05.

***Experiment 3 – Saline-alkaline condition.*** ‘Barnes’ and ‘A11’ seedlings were exposed to a 2 levels of sulfate salinity (Na2SO4:MgSO4, 1:1 molar ratio, at 6 or 12 dS m-1) x 2 alkalinity levels (NaHCO3:Na2CO3 at 5 mM at 6:4 or 2:8 molar ratio). Plants were grown under the stress for 3 weeks. The experimental design was a split-plot design, with the combination of salt type x salinity level being the whole-plot treatment arranged in a randomized complete block design with three replicates, and cultivar being the sub-plot treatment. A control treatment (i.e. non-stress) was also included in the experiment.

The two cultivars showed no differences in growth habits, such as tissue biomass and root length, under the control treatment (Table 4). No two-way or three-way interactions were observed under the stress (data not shown). Cultivar differences were only observed in shoot dry weight under the stress. Shoot water content was lower at 12 dS m-1 than at 6 dS m-1. Higher alkalinity resulted in lower shoot and root biomass, absolute water content, and root water content.

Table 4. Soybean seedling growth under the non-stress and saline-alkali stress conditions.

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
|  | Shoot dry weight(g) | Root dry weight(g) | Root length(cm) | Absolute water content(g) | Relative shoot water content(%) | Relative root water content(%) |
| Non-stress condition |
| A11 |  3.04ay | 0.53a | 59.1a | 14.3a | 82.7a | 93.4a |
| Barnes | 3.92a | 0.60a | 60.8a | 18.9a | 83.5a | 93.3a |
| Saline-alkali condition |
| Cultivar  |
| A11 | 0.60b | 0.14a | 30.4a | 2.3a | 76.7a | 91.5a |
| Bareness | 0.73a | 0.17a | 29.5a | 2.4a | 74.9a | 90.8a |
| Salinity (dS m-1)z |
| 6 | 0.72a | 0.17a | 29.2a | 2.6a | 77.8a | 91.1a |
| 12 | 0.61a | 0.14a | 30.7a | 2.1a | 73.7b | 91.1a |
| Alkalinityz |
| 6:4 | 0.76a | 0.18a | 30.8a | 2.7a | 75.8a | 91.6a |
| 2:8 | 0.57b | 0.13b | 29.1a | 2.0b | 75.7a | 90.6b |

zSalinity and alkalinity was induced by Na2SO4:MgSO4 mixtures (1:1, molar ratio) at 6 or 12 dS/m and NaHCO3:Na2CO3 mixtures at 5 mM at 6:4 or 2:8 molar ratio, respectively.

yMeans followed by the same letter within the non-stress or saline-alkali condition are not significantly different at *P* ≤ 0.05.

No cultivar differences were observed in plant nutrients under the control treatment, except Zn (1.7 mg/100 g dry weight for ‘A11’ vs. 1.5 mg/100 g dry weight for ‘Barnes’) (data not shown). Tissue nutrient levels were affected by the main factors, cultivar, salinity, and alkalinity. No two-way or three-way interactions were observed. ‘A11’ had a higher content than ‘Barnes’ in all elements, except Na (Table 5). Salinity increased tissue Na and Zn levels, but decreased Ca content. Alkalinity increased Na, Mn, and Zn uptake.

Table 5. Shoot nutrient contents (mg/100 g dry weight) of soybean seedlings grown under saline-alkalinity condition in a hydroponic system.

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
|  | K | Na | Ca | Fe | Mn | Zn |
| Cultivar |  |  |  |  |  |  |
| A11 | 1829.6a | 1648.9a |  459.3a |  7.3a |  6.0a |  0.9a |
| Barnes | 1605.7b | 1379.9a |  356.6b |  5.2b |  4.0b |  0.8b |
| Na2SO4 : MgSO4 (1:1 molar ratio) (dS m-1) |
| 6 | 1739.0a | 1042.8b |  464.4a |  5.8a |  5.2a | 0.7b |
| 12 | 1696.3a | 1985.9a |  351.5b |  6.7a |  4.8a | 0.9a |
| NaHCO3:Na2CO3 (5 mM, molar ratioio)  |
| 6:4 | 1720.7a | 1101.8b |  427.9a |  6.8a |  6.4a | 0.7b |
| 2:8 | 1714.6a | 1927.0a |  388.1a |  5.7a |  3.6b | 0.9a |

zMeans followed by the same letter within the same main factor in a nutrient are not significantly different at *P* ≤ 0.05.

1. **Evaluating tolerance of 50 soybean genotypes to the aforementioned three stresses**

Fifty genotypes were used in this study, including 9 glyphosate-resistant and 41 non-resistant. The resistant and non-resistant genotypes were analyzed separately. Seeds of each genotype were germinated and acclimated to the hydroponic system as described previously and then exposed to a Na2SO4:MgSO4 mixture (1:1, molar ratio) at 8 dS m-1 for 14 days (salinity – Experiment 4), a NaHCO3:Na2CO3 (5 mM) (2:8, molar ratio) for 21 days (alkalinity – Experiment 5), or a combination of Na2SO4:MgSO4 mixture (1:1, molar ratio) at 8 dS m-1 + NaHCO3:Na2CO3 (5 mM) (2:8, molar) for 10 days (saline-alkalinity – Experiment 6). The experimental design was a split-plot design for all three experiments, with the growing condition (non-stress vs. stress) being the whole-plot treatment arranged in a randomized complete block design with three replicates, and genotype being the sub-plot treatment. Data were collected on shoot and root dry weight, root length, absolute water content, shoot and root relative water content, and chlorophyll content. Leaf chlorosis was rated visually with a 1 – 5 scale, in which 1 = no leaf chlorosis and 5 = the worst leaf chlorosis, under alkalinity and saline-alkalinity. No leaf chlorosis was observed in plants under the non-stressful condition. Performance index indicates the number of times a genotype is ranked in the top statistical group in the variables that statistical differences are observed.

***Experiment 4 - Salinity.*** Limited differences were observed in the glyphosate-resistant group when exposed to sulfate-salinity (Table 6). In contrast, salinity resulted in reduced shoot dry weight, absolute water content, and relative shoot water content, and chlorophyll content in the non-resistant group (Table 7). ‘ND-13-15647 (CHK)’, ‘ND-13-7510 (TOFU)’, ‘ND-14-4507 (SCN), ‘ND-15-18643’, ‘ND-15-19597’, ‘ND-15-20261’, ‘ND-15-18939’, ‘ND-15-19711’, and ‘Barnes’ performed relatively better (performance index ≥ 4) (Table 7).

***Experiment 5 - Alkalinity.*** Alkalinity inhibited growth of both glyphosate-resistant and non-resistant plants (Tables 8 and 9). Genotypic differences were observed in shoot and root biomass and absolute water content in both groups and in relative root water content in the resistant group when data were pooled across stress. No significant genotypic differences were observed in leaf chlorophyll content and leaf chlorosis. Genotypes with relative high overall performance were ‘ND-15-20611’, ‘ND-15-20563’, ‘ND-15-22872’, ‘ND-15-22860’, and ‘ND-15-20592’ in the resistant group (Table 8). And 18 genotypes in the non-resistant group had performance index ≥ 2 (Table 9).

***Experiment 6 – Saline-alkalinity.*** Stress resulted in lower tissue biomass, absolute water content, and root length (non-resistant group only) (Tables 10 and 11). Similarly to Experiment 4, no genetic differences were observed in chlorophyll content and leaf damage in the stressed plants. ‘ND-15-20592’ and ‘Barnes’ had the highest tissue dry weight and absolute water content in the resistant and non-resistant group, respectively, when data were pooled across stress.

**General conclusions:**

1. Salinity, alkalinity, and the combined saline-alkalinity inhibited soybean seedling growth. Higher damage was more likely to be seen under the combined stress than individual stress.
2. NaCl was more detrimental to soybean plants than the sulfate-salinity.
3. Genetic differences were mostly observed in growth indices. Limited differences were detected in leaf chlorophyll content and chlorosis.
4. Genotypes recommended for the combined saline-alkaline condition may be different from those for individual salinity and alkalinity condition.

Table 6. Seedling growth of glyphosate-resistant soybean as affected by salinity and genotype in a hydroponic system. Salinity was induced by a Na2SO4:MgO4 mixture (1:1, molar ratio) at 8 dS m-1 for 14 days. Performance index indicates the number of times a genotype is ranked in the top statistical group in the variables that statistical differences are observed. Data were pooled across genotype or stress.

|  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
| Treatment  | Shoot dry weight(g) | Root dry weight(g) | Root length(cm) | Absolute water content(g) | Relative shootwater content (%) | Relative root water content(%) | Chlorophyll content | Performance index(Max. = 1) |
| Stress |  |  |  |  |  |  |  |  |
| Non-stress | 0.810az | 0.138a | 39.1a | 4.706a | 84.6a | 93.0a |  32.8a |  |
| Saline | 0.637a | 0.142a | 31.1a | 2.998a | 79.7b | 94.6a |  28.5b |  |
| Genotype |  |  |  |  |  |  |  |  |
| ND-15-20611  | 0.997a | 0.212a | 38.8a | 5.084a | 83.9a | 94.1a |  34.0a | 1 |
| ND-15-20563  | 0.766a | 0.129a | 34.5a | 4.106a | 77.0a | 94.4a |  34.0a | 1 |
| ND-15-20491  | 0.723a | 0.135a | 40.4a | 4.150a | 84.7a | 94.5a |  31.4ab | 1 |
| ND-14-6284  | 0.549a | 0.119a | 34.7a | 2.868a | 83.8a | 94.0a |  30.6ab | 1 |
| ND-15-20671  | 0.608a | 0.101a | 29.5a | 3.150a | 78.8a | 94.4a |  20.2c | 0 |
| ND-15-22872  | 0.788a | 0.171a | 37.8a | 4.228a | 83.9a | 92.1a |  32.3ab | 1 |
| ND-15-22860  | 0.972a | 0.184a | 36.1a | 5.250a | 84.4a | 94.2a |  33.8a | 1 |
| ND-15-22226  | 0.738a | 0.149a | 37.6a | 4.183a | 85.4a | 91.1a |  32.3ab | 1 |
| ND-15-20592  | 0.372a | 0.059a | 26.4a | 1.650a | 76.8a | 95.2a |  27.2bc | 0 |

zMeans followed by the same letter within the same factor in a column are not significantly different at *P* ≤ 0.05.

Table 7. Seedling growth of non-glyphosate-resistant soybean as affected by salinity and genotype in a hydroponic system. Salinity was induced by a Na2SO4:MgSO4 mixture (1:1, molar ratio) at 8 dS m-1 for 14 days. Performance index indicates the number of times a genotype is ranked in the top statistical group in the variables that statistical differences are observed. Data were pooled across genotype or stress.

|  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
|  | Shoot dry weight(g) | Root dry weight(g) | Root length(cm) | Absolute water content(g) | Relative shootwater content (%) | Relative root water content(%) | Chlorophyll content | Performance index(Max. = 5) |
| Stress |  |  |  |  |  |  |  |  |
| Non-stress | 0.847 az | 0.141a | 42.1a | 4.978a | 85.4a | 93.8a | 33.0a |  |
| Saline | 0.403b | 0.099a | 34.0a | 1.939b | 80.9b | 93.9a | 25.5b |  |
| Cultivar |  |  |  |  |  |  |  |  |
| ND-13-15647 (CHK) | 1.158a | 0.218ab | 36.9a | 5.960a | 83.7a | 93.6a-c | 36.0a | 5 |
| ND-13-4508 | 0.750a-f | 0.126c-h | 37.5a | 4.101a-i | 81.3a | 93.8a-c | 29.3b-e | 3 |
| ND-14-2194 (SCN) | 0.527e-g | 0.084gh | 33.2a | 2.870d-j | 84.6a | 94.8ab | 32.2a-c | 2 |
| ND-14-2671 (SCN) | 0.631b-g | 0.112d-h | 43.3a | 3.945a-i | 86.3a | 95.4a | 31.6a-d | 3 |
| ND-14-3606 (SCN) | 0.590b-g | 0.099e-h | 36.7a | 3.344b-j | 84.5a | 94.9ab | 24.8ef | 1 |
| ND-13-7510 (TOFU) | 0.814a-f | 0.149a-h | 42.7a | 4.468a-i | 84.6a | 94.2a-c | 27.4c-f | 4 |
| ND-13-4810 (TOFU) | 0.479e-g | 0.098e-h | 29.3a | 2.755d-j | 84.4a | 94.1a-c | 29.1b-e | 1 |
| ND-11-19938 (NATTO) | 0.222g | 0.056h | 25.1a | 1.228j | 82.7a | 94.6ab | 27.8c-f | 1 |
| ND-12-20324 (NATTO) | 0.448e-g | 0.089f-h | 35.4a | 2.711d-j | 85.8a | 93.9a-c | 28.6b-e | 1 |
| ND-14-4327 | 0.523e-g | 0.100e-h | 37.3a | 2.500e-j | 83.2a | 94.9ab | 30.9a-e | 2 |
| ND-14-4598 | 0.543c-g | 0.130c-h | 41.4a | 3.061d-j | 84.4a | 93.8a-c | 31.4a-e | 2 |
| ND-15-17909 | 0.812a-f | 0.185a-e | 39.8a | 4.505a-h | 85.0a | 93.9a-c | 25.7d-f | 4 |
| ND-15-18005 | 0.571c-g | 0.133c-h | 42.6a | 2.939d-j | 79.4a | 94.2a-c | 32.1a-d | 2 |
| ND-15-18034 | 0.538d-g | 0.118c-h | 33.5a | 2.804d-j | 83.4a | 94.0a-c | 31.2a-e | 2 |
| ND-15-18237 | 0.819a-f | 0.135b-h | 42.0a | 4.434a-i | 83.9a | 94.3a- | 28.5c-e | 3 |
| ND-15-17728 | 0.367fg | 0.065h | 32.8a | 1.773ij | 76.0a | 94.9ab | 20.6f | 1 |
| ND-14-4507 (SCN) | 0.831a-f | 0.110d-h | 34.8a | 4.686a-g | 82.6a | 95.3a | 31.5a-d | 4 |
| ND-15-18076 | 0.465e-g | 0.100e-h | 38.4a | 2.852d-j | 85.3a | 93.8a-c | 26.0c-f | 1 |
| ND-15-18097 | 0.493e-g | 0.102e-h | 38.9a | 2.532e-j | 80.8a | 94.5a-c | 21.2f | 1 |
| ND-15-18098 | 0.670b-g | 0.132c-h | 38.4a | 3.631a-j | 79.7a | 93.7a- | 30.0a-e | 3 |
| ND-15-18230 | 0.565-g | 0.094f-h | 31.8a | 3.454a-j | 85.8a | 94.8ab | 30.6a-e | 3 |
| ND-14-3926 (SCN) | 0.423e-g | 0.078h | 33.1a | 2.550e-j | 84.8a | 94.0a-c | 35.9ab | 2 |
| ND-15-18643 | 0.774a-f | 0.227a | 40.9a | 4.626a-h | 84.8a | 93.3a-c | 31.6a-d | 5 |
| ND-15-19597 | 1.015a-c | 0.198a-c | 41.7a | 5.725a-c | 83.8a | 93.9a-c | 31.5a-d | 5 |
| ND-15-20352 | 0.376fg | 0.066h | 30.0a | 2.036h-j | 81.3a | 94.2a-c | 30.8a-e | 2 |
| ND-15-20392 | 0.851a-e | 0.131c-h | 40.1a | 4.628a-h | 83.8a | 94.3a-c | 29.6b-e | 3 |
| ND-15-19289 | 0.713a-f | 0.107d-h | 33.9a | 3.768a-j | 81.3a | 92.8a-d | 27.0c-f | 3 |
| ND-15-18457 | 0.427e-g | 0.103e-h | 40.3a | 2.286f-j | 81.7a | 93.1a-c | 28.8b-e | 2 |
| ND-15-18596 | 0.430e-g | 0.115c-h | 61.3a | 2.227f-j | 83.9a | 87.4e | 28.2c-f | 0 |
| ND-15-19739 | 0.503e-g | 0.098e-h | 38.3a | 2.918d-j | 84.2a | 93.7a-c | 25.6d-f | 1 |
| ND-15-20261 | 0.840a-e | 0.174a-f | 41.9a | 4.783a-f | 84.7a | 91.5cd | 30.2a-e | 4 |
| ND-15-19239 | 0.562c-g | 0.100e-h | 37.5a | 2.976d-j | 83.0a | 93.8a-c | 27.4c-f | 1 |
| ND-14-2678 (SCN) | 0.558c-g | 0.104d-h | 36.5a | 3.110c-j | 83.1a | 94.6a-c | 25.8d-f | 1 |
| ND-15-18939 | 1.006a-d | 0.187a-e | 44.8a | 5.444a-d | 82.6a | 92.2b-d | 33.4a-c | 4 |
| ND-15-19711 | 0.850a-e | 0.164a-g | 41.6a | 4.913a-e | 85.4a | 92.6a-d | 29.9b-e | 4 |
| ND-15-19627 | 0.421e-g | 0.091f-h | 37.5a | 2.330e-j | 82.0a | 89.4de | 25.4d-f | 0 |
| ND-15-18515 | 0.368fg | 0.059h | 34.4a | 2.058g-j | 76.6a | 93.9a-c | 30.5a-e | 2 |
| ND-15-18287 | 0.633b-g | 0.124c-h | 36.6a | 3.399b-j | 83.7a | 94.9ab | 29.4b-e | 1 |
| ND-15-20399 | 0.600b-g | 0.116c-h | 38.4a | 3.095c-j | 82.6a | 94.1a-c | 29.4b-e | 1 |
| A11 | 0.402e-g | 0.068h | 35.2a | 2.490e-j | 84.8a | 94.7ab | 29.6b-e | 1 |
| Barnes | 1.055ab | 0.195a-d | 46.2a | 5.898ab | 84.2a | 94.5a-c | 32.0a-d | 5 |

zMeans followed by the same letter within the same factor in a column are not significantly different at *P* ≤ 0.05.

Table 8. Seedling growth of glyphosate-resistant soybean as affected by alkalinity and genotype in a hydroponic system. Alkalinity was induced by a NaHCO3:Na2CO3 (5 mM) (2:8, molar ratio) for 21 days. Leaf chlorosis was rated with a 1 – 5 score (1 = no leaf damage and 5 = the most damage) only in the stressed plants because no leaf damage was observed in non-stressed plants. Performance index indicates the number of times a genotype is ranked in the top statistical group in the variables that statistical differences are observed. Data were pooled across genotype or stress.

|  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| Treatment  | Shoot dry weight(g) | Root dry weight(g) | Root length(cm) | Absolute water content(g) | Relative shootwater content (%) | Relative root water content(%) | Chlorophyll content | Leaf chlorosis | Performance index(Max. = 4) |
| Stress |  |  |  |  |  |  |  |  |  |
| Non-alkalinity |  1.052az |  0.158a | 42.3a |  5.736a | 85.1a |  93.1a | 27.4a |  |  |
| Alkalinity |  0.427b |  0.080b | 18.1b |  1.489b | 77.9b |  90.1b | 15.8b |  |  |
| Genotype |  |  |  |  |  |  |  |  |  |
| ND-15-20611  |  0.880ab |  0.163ab | 33.0a |  4.275ab | 81.0a |  91.2bc | 23.1a | 2.9a | 3 |
| ND-15-20563  |  0.838ab |  0.129a-c | 32.5a |  3.895a-d | 80.9a |  91.3bc | 22.8a | 2.6a | 3 |
| ND-15-20491  |  0.456cd |  0.046d | 26.3a |  2.264cd | 82.1a |  93.3a | 21.9a | 3.4a | 1 |
| ND-14-6284  |  0.643b-d |  0.103b-d | 32.3a |  3.624a-d | 82.8a |  89.6c | 21.1a | 3.1a | 1 |
| ND-15-20671  |  0.607b-d |  0.106b-d | 27.0a |  3.060b-d | 82.5a |  91.7ab | 18.7a | 3.7a | 1 |
| ND-15-22872  |  0.828a-c |  0.131a-c | 26.3a |  4.129a-c | 81.7a |  92.4ab | 22.4a | 2.8a | 4 |
| ND-15-22860  |  0.805a-d |  0.140a-d | 30.7a |  3.976a-d | 81.6a |  91.5b | 19.7a | 3.5a | 3 |
| ND-15-22226  |  0.419d |  0.070cd | 26.6a |  2.081d | 82.5a |  92.2ab | 21.1a | 3.1a | 1 |
| ND-15-20592  |  1.182a |  0.184a | 37.4a |  5.207a | 79.4a |  91.7ab | 23.8a | 2.6a | 4 |

zMeans followed by the same letter within the same factor in a column are not significantly different at *P* ≤ 0.05.

Table 9. Seedling growth of non-glyphosate-resistant soybean as affected by alkalinity and genotype in a hydroponic system. Alkalinity was induced by a NaHCO3:Na2CO3 (5 mM) (2:8, molar ratio) for 21 days. Leaf chlorosis was rated with a 1 – 5 score (1 = no leaf damage and 5 = the most damage) only in the stressed plants because no leaf damage was observed in non-stressed plants. Performance index indicates the number of times a genotype is ranked in the top statistical group in the variables that statistical differences are observed. Data were pooled across genotype or stress.

|  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| Treatment | Shoot dry weight(g) | Root dry weight(g) | Root length(cm) | Absolute water content(g) | Relative shootwater content (%) | Relative root water content(%) | Chlorophyll content | Leaf chlorosis | Performance index(Max. = 3) |
| Stress |  |  |  |  |  |  |  |  |  |
| Non-stress | 1.073a | 0.163a | 45.0a | 6.090a | 85.4a | 93.8a | 28.8a |  |  |
| Saline | 0.462b | 0.090b | 22.2b | 1.579b | 77.5b | 90.2b | 14.2b |  |  |
| Cultivar |  |  |  |  |  |  |  |  |  |
| ND-13-15647 (CHK) | 0.984a-e | 0.156a-d | 35.4a | 4.528a-d | 81.1a | 92.2a | 19.7a | 2.7a | 3 |
| ND-13-4508 | 0.747c-h | 0.099c-h | 29.5a | 3.954a-f | 82.1a | 92.5a | 17.6a | 3.3a | 1 |
| ND-14-2194 (SCN) | 0.864a-g | 0.182ab | 31.4a | 3.482b-g | 80.9a | 91.1a | 23.1a | 2.5a | 2 |
| ND-14-2671 (SCN) | 0.676c-i | 0.126b-h | 40.2a | 3.709b-g | 83.0a | 92.1a | 20.4a | 3.0a | 0 |
| ND-14-3606 (SCN) | 0.774b-h | 0.120b-h | 31.1a | 3.876b-f | 81.8a | 92.4a | 19.9a | 3.0a | 0 |
| ND-13-7510 (TOFU) | 0.637e-i | 0.090d-h | 34.0a | 3.411b-g | 81.7a | 92.0a | 22.4a | 3.2a | 0 |
| ND-13-4810 (TOFU) | 0.866a-g | 0.133b-g | 35.8a | 4.396a-d | 81.1a | 92.4a | 21.2a | 3.8a | 2 |
| ND-11-19938 (NATTO) | 0.342i | 0.059h | 27.7a | 1.794g | 82.7a | 91.1a | 22.2a | 2.9a | 0 |
| ND-12-20324 (NATTO) | 0.709c-i | 0.098c-h | 34.8a | 3.983a-f | 82.3a | 92.3a | 18.3a | 3.8a | 1 |
| ND-14-4327 | 0.919a-g | 0.175ab | 34.4a | 4.799a-c | 82.6a | 91.3a | 20.7a | 3.8a | 3 |
| ND-14-4598 | 0.920a-g | 0.179ab | 31.9a | 4.470a-d | 81.4a | 91.2a | 18.9a | 3.7a | 3 |
| ND-15-17909 | 0.663c-i | 0.099c-h | 33.2a | 2.985d-g | 79.9a | 91.7a | 21.9a | 3.4a | 0 |
| ND-15-18005 | 0.786b-h | 0.155a-d | 37.8a | 4.114a-f | 82.1a | 91.6a | 18.6a | 3.8a | 2 |
| ND-15-18034 | 0.710c-i | 0.129b-h | 39.0a | 3.241b-g | 80.2a | 91.7a | 22.8a | 2.8a | 0 |
| ND-15-18237 | 0.897a-g | 0.143a-f | 36.7a | 4.545a-d | 81.9a | 92.7a | 21.5a | 2.9a | 3 |
| ND-15-17728 | 0.825a-h | 0.163a-c | 37.3a | 4.018a-f | 80.6a | 92.4a | 24.7a | 2.9a | 3 |
| ND-14-4507 (SCN) | 0.757c-h | 0.149a-e | 36.5a | 3.949a-f | 83.2a | 90.5a | 21.1a | 3.1a | 2 |
| ND-15-18076 | 0.774b-h | 0.128b-h | 35.7a | 3.944a-f | 81.0a | 92.2a | 22.9a | 3.3a | 1 |
| ND-15-18097 | 1.002a-d | 0.168a-c | 34.2a | 4.561a-d | 79.5a | 91.9a | 20.4a | 3.8a | 3 |
| ND-15-18098 | 0.773b-h | 0.132b-h | 39.8a | 3.756b-f | 81.1a | 91.9a | 20.3a | 3.4a | 0 |
| ND-15-18230 | 0.833a-h | 0.138a-g | 34.8a | 4.856ab | 82.1a | 92.9a | 20.0a | 3.9a | 3 |
| ND-14-3926 (SCN) | 0.683c-i | 0.096d-h | 30.2a | 3.433b-g | 82.4a | 92.5a | 21.5a | 3.3a | 0 |
| ND-15-18643 | 0.662c-i | 0.081f-h | 29.9a | 3.159b-g | 80.8a | 92.3a | 18.7a | 3.8a | 0 |
| ND-15-19597 | 1.016a-c | 0.167a-c | 34.2a | 4.784a-c | 80.8a | 92.8a | 21.5a | 3.0a | 3 |
| ND-15-20352 | 0.490hi | 0.078f-h | 30.3a | 2.503e-g | 83.1a | 92.7a | 22.7a | 2.9a | 0 |
| ND-15-20392 | 0.693c-i | 0.103c-h | 29.3a | 3.203b-g | 81.5a | 92.4a | 20.4a | 2.8a | 0 |
| ND-15-19289 | 0.625f-i | 0.089d-h | 29.9a | 2.951d-g | 80.5a | 91.7a | 21.5a | 3.0a | 0 |
| ND-15-18457 | 0.714c-i | 0.146a-f | 32.9a | 3.406b-g | 81.0a | 91.8a | 22.2a | 2.8a | 1 |
| ND-15-18596 | 0.492hi | 0.073gh | 27.2a | 2.404fg | 82.0a | 92.4a | 22.5a | 2.8a | 0 |
| ND-15-19739 | 0.807a-h | 0.122b-h | 37.1a | 4.325a-d | 82.0a | 92.2a | 26.0a | 2.8a | 2 |
| ND-15-20261 | 0.741c-h | 0.112b-h | 32.8a | 3.824b-f | 82.2a | 92.8a | 20.8a | 3.4a | 0 |
| ND-15-19239 | 0.621f-i | 0.074gh | 27.0a | 3.022c-g | 80.9a | 92.0a | 26.7a | 4.2a | 0 |
| ND-14-2678 (SCN) | 0.955a-f | 0.164a-c | 34.3a | 4.806ab | 82.1a | 92.5a | 22.9a | 2.3a | 3 |
| ND-15-18939 | 1.152ab | 0.171a-c | 42.2a | 5.810a | 81.3a | 92.6a | 23.7a | 2.6a | 3 |
| ND-15-19711 | 0.802a-h | 0.131b-h | 33.4a | 4.255a-e | 82.5a | 92.3a | 21.1a | 3.5a | 2 |
| ND-15-19627 | 0.632e-i | 0.081e-h | 26.3a | 3.340b-g | 81.8a | 92.1a | 21.7a | 3.2a | 0 |
| ND-15-18515 | 0.795a-h | 0.122b-h | 33.6a | 4.211a-f | 81.3a | 91.6a | 18.2a | 4.1a | 2 |
| ND-15-18287 | 0.591g-i | 0.123b-h | 33.6a | 2.884d-g | 81.2a | 92.3a | 20.2a | 3.8a | 0 |
| ND-15-20399 | 0.729c-i | 0.117b-h | 30.2a | 3.452b-g | 80.8a | 92.2a | 24.0a | 2.6a | 0 |
| A11 | 0.649d-i | 0.121b-h | 32.9a | 3.273b-g | 83.0a | 92.3a | 22.2a | 2.4a | 0 |
| Barnes | 1.159a | 0.203a | 39.2a | 5.798a | 81.9a | 91.9a | 22.8a | 3.3a | 3 |

zMeans followed by the same letter within the same factor in a column are not significantly different at *P* ≤ 0.05.

Table 10. Seedling growth of glyphosate-resistant soybean as affected by saline-alkalinity and genotype in a hydroponic system. Salinity was induced by a combination of Na2SO4:MgSO4 mixture (1:1, molar ratio) at 8 dS m-1 and NaHCO3:Na2CO3 (5 mM) (2:8, molar ratio) for 10 days. Leaf chlorosis was rated with a 1 – 5 score (1 = no leaf damage and 5 = the most damage) only in the stressed plants because no leaf damage was observed in non-stressed plants. Performance index indicates the number of times a genotype is ranked in the top statistical group in the variables that statistical differences are observed. Data were pooled across genotype or stress.

|  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| Treatment  | Shoot dry weight(g) | Root dry weight(g) | Root length(cm) | Absolute water content(g) | Relative shootwater content (%) | Relative root water content(%) | Chlorophyll content | Leaf chlorosis | Performance index(Max. = 3) |
| Stress |  |  |  |  |  |  |  |  |  |
| Non-stress | 0.383a | 0.062a | 39.8a | 2.549a | 87.4a | 94.2a | 28.9a |  |  |
| Saline-alkali | 0.251b | 0.035b | 25.8b | 1.109b | 81.7b | 92.3b | 17.7b |  |  |
| Genotype |  |  |  |  |  |  |  |  |  |
| ND-15-20611  | 0.330bc | 0.050b | 28.3a | 2.031bc | 85.5a | 93.6a | 27.2a | 1.5a | 0 |
| ND-15-20563  | 0.288bc | 0.043b | 31.4a | 1.824bc | 84.7a | 91.3a | 20.7a | 2.0a | 0 |
| ND-15-20491  | 0.268bc | 0.038b | 29.8a | 1.479bc | 83.6a | 94.2a | 22.0a | 4.0a | 0 |
| ND-14-6284  | 0.165c | 0.027b | 34.5a | 1.190c | 88.1a | 93.4a | 24.6a | 2.0a | 0 |
| ND-15-20671  | 0.218bc | 0.031b | 30.5a | 1.252c | 83.3a | 94.7a | 22.5a | 3.0a | 0 |
| ND-15-22872  | 0.366b | 0.058b | 36.8a | 2.199b | 84.9a | 93.7a | 23.0a | 2.7a | 0 |
| ND-15-22860  | 0.278bc | 0.026b | 25.1a | 1.467bc | 84.6a | 93.8a | 23.8a | 2.5a | 0 |
| ND-15-22226  | 0.339bc | 0.068ab | 36.5a | 1.823bc | 83.9a | 91.4a | 24.5a | 3.5a | 1 |
| ND-15-20592  | 0.601a | 0.094a | 42.5a | 3.983a | 83.1a | 93.1a | 21.4a | 3.5a | 3 |

zMeans followed by the same letter within the same factor in a column are not significantly different at *P* ≤ 0.05.

Table 11. Seedling growth of non-glyphosate-resistant soybean as affected by saline-alkalinity and genotype in a hydroponic system. Salinity was induced by a combination of Na2SO4:MgSO4 mixture (1:1, M:M) at 12 dS m-1 + NaHCO3:Na2CO3 (5 mM) (2:8, M:M ratio) for 10 days. Leaf chlorosis was only rated under alkalinity as no leaf damage was observed in non-stressed plants. Performance index indicates the number of times a genotype is ranked in the top statistical group in the variables that statistical differences are observed. Data were pooled across genotype or stress.

|  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| Treatment | Shoot dry weight(g) | Root dry weight(g) | Root length(cm) | Absolute water content(g) | Relative shootwater content (%) | Relative root water content(%) | Chlorophyll content | Leaf chlorosis | Performance index(Max. = 4) |
| Stress |  |  |  |  |  |  |  |  |  |
| Non-stress | 0.433a | 0.065a | 44.1a | 2.770a | 86.3a | 94.6a | 30.3a |  |  |
| Saline-alkali | 0.269b | 0.036b | 25.4b | 1.141b | 81.1b | 92.4b | 17.4b |  |  |
| Cultivar |  |  |  |  |  |  |  |  |  |
| ND-13-15647 (CHK) | 0.440bc | 0.067bc | 37.8b-d | 2.462b-f | 83.8a | 93.4a | 29.0a | 2.0a | 0 |
| ND-13-4508 | 0.312c-e | 0.040b-e | 40.4b-d | 1.747c-j | 84.6a | 94.1a | 25.8a | 2.0a | 0 |
| ND-14-2194 (SCN) | 0.386b-d | 0.047b-e | 38.8b-d | 1.895c-f | 82.3a | 94.3a | 23.8a | 3.2a | 0 |
| ND-14-2671 (SCN) | 0.166e | 0.027de | 23.5f | 1.044h-j | 84.6a | 92.5a | 21.9a | 3.0a | 0 |
| ND-14-3606 (SCN) | 0.185e | 0.020e | 25.2f | 1.037ij | 84.5a | 95.6a | 18.9a | 2.2a | 0 |
| ND-13-7510 (TOFU) | 0.446bc | 0.065b-d | 40.0 | 2.441b-g | 83.2a | 93.6a | 23.4a | 2.8a | 0 |
| ND-13-4810 (TOFU) | 0.169e | 0.022e | 25.2b-d | 0.778j | 82.2a | 93.5a | 25.9a | 1.5a | 0 |
| ND-11-19938 (NATTO) | 0.271c-e | 0.046b-e | 35.5f | 1.490f-j | 84.2a | 93.5a | 21.6a | 2.2a | 0 |
| ND-12-20324 (NATTO) | 0.321c-e | 0.040b-e | 30.4d-f | 1.669d-j | 83.6a | 93.7a | 26.5a | 3.5a | 0 |
| ND-14-4327 | 0.422b-d | 0.066bc | 40.8b-d | 2.494b-e | 83.8a | 94.2a | 25.3a | 3.2a | 0 |
| ND-14-4598 | 0.322c-e | 0.050b-e | 32.7c-f | 1.769c-j | 81.7a | 94.0a | 21.5a | 3.5a | 0 |
| ND-15-17909 | 0.426b-d | 0.062b-d | 41.8a-d | 2.344b-h | 83.5a | 93.8a | 23.5a | 2.8a | 1 |
| ND-15-18005 | 0.431b-d | 0.067bc | 47.8ab | 2.228b-h | 82.4a | 93.1a | 23.8a | 3.0a | 1 |
| ND-15-18034 | 0.510b | 0.068b | 36.7c-e | 2.554b-d | 82.6a | 93.5a | 29.3a | 2.0a | 0 |
| ND-15-18237 | 0.287c-e | 0.043b-e | 37.7b-e | 1.519e-j | 84.2a | 93.4a | 22.0a | 3.0a | 0 |
| ND-15-17728 | 0.320c-e | 0.046b-e | 35.2c-f | 1.651d-j | 82.3a | 94.6a | 17.8a | 3.3a | 0 |
| ND-14-4507 (SCN) | 0.493b | 0.066bc | 39.8b-d | 3.095b | 84.7a | 94.3a | 25.0a | 1.8a | 0 |
| ND-15-18076 | 0.248de | 0.035de | 28.0ef | 1.421g-j | 82.5a | 91.5a | 19.4a | 1.3a | 0 |
| ND-15-18097 | 0.304c-e | 0.040b-e | 34.5c-f | 1.688d-j | 84.6a | 94.1a | 23.6a | 2.8a | 0 |
| ND-15-18098 | 0.311c-e | 0.036de | 32.2c-f | 1.611d-j | 83.9a | 93.3a | 21.8a | 2.3a | 0 |
| ND-15-18230 | 0.347b-e | 0.065b-d | 35.0c-f | 2.115b-i | 86.0a | 93.5a | 24.0a | 3.5a | 0 |
| ND-14-3926 (SCN) | 0.300c-e | 0.051b-e | 30.9d-f | 1.483g-j | 82.6a | 93.9a | 25.5a | 2.2a | 0 |
| ND-15-18643 | 0.411b-d | 0.061b-d | 42.0a-c | 2.161b-h | 83.1a | 93.7a | 22.2a | 3.8a | 1 |
| ND-15-19597 | 0.329c-e | 0.037de | 34.8c-f | 1.921c-j | 84.2a | 93.4a | 25.0a | 2.3a | 0 |
| ND-15-20352 | 0.253c-e | 0.029de | 23.5f | 1.482g-j | 84.5a | 94.4a | 27.6a | 4.0a | 0 |
| ND-15-20392 | 0.436bc | 0.061b-d | 37.6b-e | 2.755bc | 83.7a | 92.8a | 26.3a | 2.2a | 0 |
| ND-15-19289 | 0.320c-e | 0.043b-e | 32.4c-f | 1.540d-j | 82.6a | 92.6a | 27.1a | 2.0a | 0 |
| ND-15-18457 | 0.351b-d | 0.060b-d | 51.3a | 2.084c-i | 85.2a | 94.4a | 20.1a | 4.0a | 1 |
| ND-15-18596 | 0.333c-e | 0.045b-e | 30.1d-f | 1.744d-j | 83.0a | 92.0a | 21.1a | 3.5a | 0 |
| ND-15-19739 | 0.443bc | 0.057b-d | 37.8b-d | 2.332b-h | 83.2a | 92.7a | 22.0a | 3.2a | 0 |
| ND-15-20261 | 0.254c-e | 0.038c-e | 32.2d-f | 1.666d-j | 86.3a | 92.6a | 21.1a | 2.5a | 0 |
| ND-15-19239 | 0.382b-d | 0.048b-e | 38.2b-d | 1.972c-i | 82.9a | 93.3a | 28.5a | 2.5a | 0 |
| ND-14-2678 (SCN) | 0.403b-d | 0.058b-d | 31.4d-f | 2.731b-d | 86.2a | 93.4a | 25.3a | 2.0a | 0 |
| ND-15-18939 | 0.303c-e | 0.033de | 23.7f | 1.911c-j | 85.7a | 94.6a | 27.0a | 2.3a | 0 |
| ND-15-19711 | 0.357b-d | 0.057b-d | 33.6c-f | 2.059c-i | 85.6a | 88.4a | 27.1a | 3.3a | 0 |
| ND-15-19627 | 0.286c-e | 0.048b-e | 34.3c-f | 1.479g-j | 82.5a | 92.3a | 24.2a | 3.7a | 0 |
| ND-15-18515 | 0.395b-d | 0.060b-d | 40.1b-d | 2.192b-h | 83.8a | 93.9a | 22.4a | 2.8a | 0 |
| ND-15-18287 | 0.289c-e | 0.052b-e | 36.5c-f | 1.542d-j | 83.6a | 94.6a | 13.4a | 3.5a | 0 |
| ND-15-20399 | 0.281c-e | 0.051b-e | 27.1ef | 1.668d-j | 83.7a | 92.7a | 26.1a | 1.8a | 0 |
| A11 | 0.431b-d | 0.049b-e | 35.5c-f | 2.279b-h | 82.3a | 94.8a | 25.4a | 2.0a | 0 |
| Barnes | 0.720a | 0.119a | 33.4c-f | 4.125a | 84.4a | 94.0a | 25.8a | 2.8a | 3 |

zMeans followed by the same letter within the same factor in a column are not significantly different at *P* ≤ 0.05.

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