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GEBZE TECHNICAL UNIVERSITY

INSTITUTE OF BIOTECHNOLOGY

PHYSIOLOGICAL INVESTIGATION OF THE RELATIONSHIP BETWEEN SALINITY AND BORON TOXICITY ON WHEAT

BURAK TUNCBAS

A THESIS SUBMITTED FOR THE DEGREE OF

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ASST. PROF. UMIT BARIS KUTMAN

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SUMMARY

Both salinity and boron (B) toxicity are among major abiotic stress factors that adversely affect crop production and thus pose a threat to global food security. Due to complex interactions between them, the effects of the combination of salinity and B toxicity on wheat crops may be different from the individual effects of these factors. To better understand this combination, bread and durum wheat species were grown with different salinity and B supply levels in soil and hydroponic culture under a controlled growth chamber and greenhouse conditions. The main effects of salinity, B supply and their interaction on vegetative growth, the distribution of selected nutrients among sink and source tissues, transpiration rate, oxidative damage, and yield parameters were determined.

While the salinity-B toxicity combination had no noticeable effect on growth, the main effects of salinity and B toxicity significantly decreased shoot and root growth, respectively. Although moderate and severe B toxicity negatively affected root growth, it was found that it did not accumulate in the root but at the tips of the leaves, and this accumulation and related necrosis and membrane damage decreased with salinity. With or without salinity, B toxicity did not contribute to the oxidative injury of the shoot, except in some cases. Boron toxicity treatments tended to reduce yield parameters, especially the grain yield and harvest index, also salinity stress reduced these parameters significantly. Further studies on other crops are needed to determine the effects of the combination of salinity and B toxicity on plants.

Key Words: Salinity stress, Boron toxicity, stress combination, bread wheat, durum wheat.

ÖZET

Hem tuzluluk hem de bor (B) toksisitesi, bitkisel üretimi olumsuz yönde etkileyen ve dolayısıyla küresel gıda güvencesine tehdit oluşturan başlıca abiyotik stres faktörleri arasındadır. Aralarındaki karmaşık etkileşim nedeniyle, tuzluluk ve B toksisitesi kombinasyonunun buğday ekinleri üzerindeki etkileri bu faktörlerin bireysel etkilerinden farklı olabilir. Bu kombinasyonu daha iyi anlamak için, ekmeklik ve makarnalık buğday türleri, farklı tuzluluk ve B seviyesi uygulanarak, toprak ve hidroponik büyüme ortamlarında, kontrollü iklimlendirme odası veya sera koşullarında yetiştirilmişlerdir. Tuzluluk ve B toksisitesi faktörlerinin yalnız başlarına ve kombinasyon oluşturarak vejetatif büyüme, seçili besin maddelerinin kaynak ve sink dokular üzerindeki dağılımı, transpirasyon hızı, oksidatif hasar ve verim parametreleri üzerindeki etkileri saptandı.

Tuzluluk ve B toksisitesi kombinasyonunun büyüme üzerinde belirgin bir etkisi olmamasına rağmen, sürgün ve kök büyümesi sırasıyla tuzluluk ve B toksisitesinin tek başlarına olan etkilerinden önemli ölçüde azalmıştır. Orta ve şiddetli B tedariği kök büyümesini olumsuz etkilemekle birlikte, kökte birikmeyip yaprak uçlarında biriktiği ve tuzlulukla birlikte bu birikim ve buna bağlı nekroz ve membran hasarının azaldığı bulunmuştur. Tuzluluk olsun ya da olmasın, bazı durumlar hariç, B toksisitesi sürgün oksidatif hasarına katkıda bulunmadı. Bor toksisitesi uygulamaları verim parametrelerini, özellikle de tane verimi ve hasat indeksini azaltma eğilimindeydi, fakat tuzluluk stresi ise bu parametreleri önemli ölçüde düşürdü. Tuzluluk ve B toksisitesi kombinasyonunun bitkiler üzerindeki etkilerini belirlemek için diğer ekinler üzerinde daha fazla çalışmaya ihtiyaç vardır.

Anahtar Kelimeler: Tuzluluk stresi, bor toksisitesi, abiyotik stress kombinasyonu, ekmeklik buğday, makarnalık buğday.

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LIST of ABBREVIATIONS and ACRONYMS

Abbreviations and Acronyms		Explanations
$(CO_3)^{-2}$	:	Carbonate
(HCO_3^-)	:	Bicarbonate
$(NH_4)6Mo_7O_{24}.4H_2O$	:	Ammonium heptamolybdate tetrahydrate
$(SO_4^-)_2$	:	Sulfate
µM	:	Micromolar
Ψ_S	:	Solute Potential
Ψ_W	:	Water potential
$^1O_2$	:	Singlet oxygen
Act	:	Activity
ANOVA	:	Analysis of variance
APX	:	Ascorbate peroxidase
ASC	:	Ascorbate
B	:	Boron
BHT	:	Butylated hydroxytoluene
BOREN	:	National Boron Institute
BS	:	Boron supply
Ca	:	Calcium
$Ca(NO_3)_2$	:	Calcium nitrate
CAT	:	Catalase
Cl^-	:	Chloride
cm	:	Centimeter
CO_2	:	Carbon dioxide
Conc.	:	Concentration
Cont.	:	Content
Cu	:	Copper
$CuSO_4.5(H_2O)$	:	Copper sulfate pentahydrate
cv.	:	cultivar
DAS	:	Days After Sowing
DF	:	Degrees of freedom

dH_2O	:	Distilled water
dS	:	Deci Siemens
DW	:	Dry weight
EC	:	Electrical Conductivity
EC_e	:	Saturated Paste Extract
ESP	:	Exchangeable Sodium Percentage
FAO	:	Food and Agriculture Organization
Fe	:	Iron
FeEDTA	:	Iron ethylenediaminetetraacetate
FW	:	Fresh weight
g	:	Gram
GB	:	Glycine Betaine
GR	:	Glutathione reductase
GSH	:	Reduced glutathione
GSSG	:	Oxidized Glutathione
h	:	Hour
H_2O	:	Water
H_2O_2	:	Hydrogen peroxide
H_3BO_3	:	Boric acid
HCl	:	Hydrochloric acid
HI	:	Harvest index
HNO_3	:	Nitric acid
HSD	:	Tukey's honestly significant difference
ICP-OES	:	Inductively coupled plasma optical emission spectrometry
K	:	Potassium
K_2SO_4	:	Potassium sulfate
KCl	:	Potassium chloride
kg	:	Kilogram
KH_2PO_4	:	Potassium dihydrogen phosphate
$KI\text{-}I_2$	:	Potassium iodide - Iodine
K-P	:	Potassium phosphate buffer
l	:	Liter
LSD	:	Fisher's least significant difference

m	:	Meter
MDA	:	Malondialdehyde
mg	:	Miligram
Mg	:	Magnesium
$MgSO_4$	:	Magnesium sulfate
min	:	Minute
ml	:	Milliliter
mM	:	Millimolar
mm	:	Millimeter
Mn	:	Manganese
$MnSO_4.H_2O$	:	Manganese sulfate monohydrate
MPa	:	Megapascal
N	:	Nitrogen
n.s.	:	not significant
Na	:	Sodium
NaCl	:	Sodium chloride
NADPH	:	Nicotinamide adenine dinucleotide phosphate
NADPH-Na_4	:	Dihydronicotinamide adenine dinucleotide phosphate tetrasodium sal
NBT	:	Nitro blue tetrazolium chloride
NH_4^+	:	Ammonium
NO_3^-	:	Nitrate
$O_2^{\cdot -}$	:	Superoxide radical
OH^-	:	Hydroxyl radical
P	:	Phosphorus
pH	:	Power of hydrogen
ppm	:	parts per million
Pr	:	Probabilities
REL	:	Relative electrolyte leakage
RG-II	:	Rhamnogalacturonan II
ROS	:	Reactive Oxygen Species
S	:	Salinity
SOD	:	Superoxide dismutase
Sp	:	Specific

TBA	:	Thiobarbituric acid
TCA	:	Trichloroacetic acid
TGW	:	Thousand grain weight
Tox	:	Toxicity
US	:	United States
UV	:	Ultraviolet
WC	:	Water content
Zn	:	Zinc
$ZnSO_4.7H_2O$	:	Zinc sulfate heptahydrate

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1. INTRODUCTION

In order to feed the growing population and ensure global food security, it is essential to increase crops yield in our world where the possibility of expanding agricultural areas is very limited and the agricultural lands currently in use are rapidly being degraded due to different causes [Godfray et al., 2010], [Tilman et al., 2011].

Abiotic stresses such as drought, salinity, freezing, heat, mineral deficiencies, boron (B) toxicity, and heavy metal toxicities are the main obstacles to the realization of the yield potentials of plants in agriculture and these abiotic stresses are expected to intensify in the future in many parts of the world as a result of global climate change as well as anthropogenic activities [Ahuja et al., 2010].

Different stress factors can affect plants individually, but under field conditions, it is common that they form a combination. The consequences of stress combinations and interactions may be very different from those of individual stresses [Mittler, 2006]. Specifically, although the interaction of salinity and B toxicity has been investigated in several studies conducted in this context, the discrepancies between the results of these studies and the gaps of knowledge about the physiological mechanisms underlying the observations are remarkable [Yermiyahu et al., 2008], [Wimmer et al., 2003], [Edelstein et al., 2005].

1.1. Abiotic Stress

Abiotic stress conditions are associated with an increase in reactive oxygen species (ROS) (*e.g.*,, $^1O_2$, $\cdot O_2^-$, $\cdot OH$) formation, causing oxidative stress in addition to their primary adverse effects on plants [Mittler, 2002]. Plants have developed various avoidance and tolerance mechanisms to cope with abiotic stress.

The antioxidative defense system scavenges ROS with enzymatic and non-enzymatic antioxidants. These antioxidative enzymes include, among others, superoxide dismutase (SOD), catalase (CAT), ascorbate peroxidase (APX), and glutathione reductase (GR) while non-enzymatic antioxidants include polyphenols, ascorbic acid, α-tocopherol, glutathione and carotenoids. Under normal conditions, various antioxidative defense mechanisms effectively scavenge the ROS molecules

and prevent oxidative damage [Gill and Tuteja, 2010] but exposure of plants to abiotic stress conditions such as salinity, freezing, drought, nutrient deficiency or heat can increase the production of ROS and consequently result in yield and quality losses in cereals and other plants.

1.2. Salinity Stress

Soil salinity, which is one of the most common major abiotic stress factors, is a serious threat that increases with anthropogenic activities, particularly in arid and semi-arid regions [Letey, 2000]. Soil salinity is a consequence of the total concentration of soluble ions in the soil and can be expressed in terms of electrical conductivity (EC) [FAO/AGL, 2018]. Important mineral salt cations and anions that affect the electrical conductivity of soil solution are: sodium (Na^+), calcium (Ca^{2+}), magnesium (Mg^{2+}), potassium (K^+), chloride (Cl^-), sulfate (SO_4^{2-}), bicarbonate (HCO_3^-), carbonate (CO_3^-), and nitrate (NO_3^-). Soil salinity is divided into two types according to its origin: primary salinity due to natural causes and secondary salinity due to human activities [Imadi et al., 2015]. Natural causes of soil salinity are geological and climatic and include low precipitation, weathering of rocks, salty groundwater and ion transportation with the wind.

The global scale of the soil affected by salinity, sodicity, and salinity-sodicity were shown in figure 1.1 [Wicke et al., 2011]. As can be seen from the map, salinity is observed in various regions of the world especially in Australia, Middle East, and North Africa. Also, soil salinity is a serious threat for central and south-east Anatolia, two of Turkey's most important agricultural lands [Kanber et al., 2005].

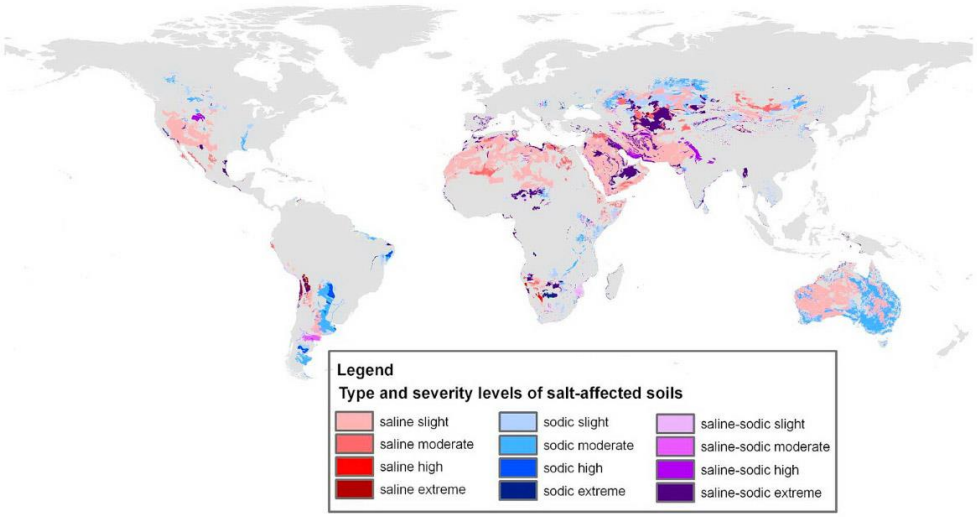

Figure 1.1: Type and severity levels of salt-affected soils.

According to the FAO data, salinity affects 20% of the total cultivated and 33% of irrigated croplands at world [FAO/AGL, 2018]. Also, soil salinity is increasing year by year due to global warming, usage of poor-quality irrigation water, and other anthropogenic activities [Shrivastava and Kumar, 2015]. Worse, it is estimated that 50 percent of the agricultural land in 2050 will suffer from salinity problems if adequate precautions are not taken.

Depending on the total concentration of soluble ions and the share of Na in total soluble ions, soils can be classified as saline, sodic or saline-sodic [Shrivastava and Kumar, 2015]. Electrical conductivity (EC) and exchangeable sodium percentage (ESP) are used to categorize soils in this context. Table 1.1 shows the differences between saline, sodic and saline-sodic soils according to US salinity laboratory.

Table 1.1 Main difference between saline, sodic and saline-sodic soils.

	Saline soils	**Sodic soils**	**Saline-sodic soils**
EC of saturated soil extract	> 4 dS (m^{-1})	< 4 dS (m^{-1})	> 4 dS (m^{-1})
Exchangeable Na (ESP) ESP (%) = {[Na]/([Ca] + [Mg] + [K] + [Na])} x 100	< 15 %	> 15 %	> 15 %
pH	< 8.5	8.5 - 10	< 8.5

The osmotic (solute) potential (Ψ_S) of the soil can be easily calculated from EC values according to the following equation:

$$\Psi_S \text{ (MPa)} = - \text{EC (dS m}^{-1}\text{)} \times 0.036$$

Increased electrical conductivity is associated with increased concentration of dissolved ions in the soil solution and reduced osmotic potential [Allbed and Kumar, 2013]. As saline soils have low osmotic potentials, water uptake from such soils becomes difficult for plant roots.

There are some limitations of just measuring EC in saturation extract to comment on soil salinity:

- No clue about salt composition
- EC of the soil solution at or below field capacity is in fact much higher.
- Rhizosphere may have a much higher EC than bulk soil.

1.2.1. Effects of Salinity on Plants

The ions of salt, sodium (Na^+) and chlorine (Cl^-), are toxic to plants above certain concentration thresholds although Cl is an essential micronutrient for higher plants while at lower concentrations, Na is considered a beneficial element for some plants. Soil salinity decreases the efficiency of agricultural activities by negatively affecting growth and yield of plants [Parvaiz and Satyawati, 2008]. In addition, seed germination, which is a critical and fragile stage of plant development, is also impaired by salinity [Läuchli and Grattan, 2007]. Depending on the salinity level, the germination of the seed is delayed and the germination percentage decreases. Loss of seedling vigor results in impaired seedling establishment. Salinity firstly affects roots, which serve as bridges between the soil and the plant [Marcar et al., 1999].

Salinity has three main physiological impacts on plants: Water deficit due to osmotic effect, specific ion toxicities, and nutrient imbalance [Läuchli and Epstein., 1990]. Also, salinity can be involved in a double or triple whammy when combined with other abiotic stress factors such as drought, B toxicity and heat [Bastías et al., 2004].

The most important driving factor behind the ability of plants to absorb water and minerals from the soil is the higher water potential in the soil [Carvajal et al., 1999]. However, it appears that salinity reduces the soil water potential and hence makes it difficult for crops to absorb water. Reduced water uptake impairs the water balance of plants. Water deficit appears, therefore, as the osmotic component of salt stress. Because of the reduced turgor in leaf cells, the elongation rate of the leaves decreases [Naz et al., 2019]. The reduction of stomatal conductance and transpiration

in plants under salt stress diminishes CO_2 uptake and leads to damage to critical physiological processes such as photosynthesis.

While the primary effect of salinity on the plant is often considered as the osmotic effect, ion toxicity is the secondary effect [Munns and Tester, 2008], [Läuchli and Grattan, 2007]. The osmotic effect of the salt on the plants is observed immediately after the salt application and the effect continues throughout the observation period [Munns, 2002]. In contrast, ionic stress, the secondary effect of salinity on plants, occurs over time as a result of the accumulation of toxic ions and causes chlorosis and necrosis in mature leaves and premature senescence of old leaves [Tester and Davenport, 2003]. The impact of the secondary effect, also called salt-specific effect, on plants is slower than osmotic stress but may even be more drastic [Läuchli and Grattan, 2007].

Chloride (Cl^-) ion, which is abundant in nature, is easily absorbed by plants and has high mobility in plant tissues [Chen et al., 2010]. While Cl deficiency is a rarely observed problem in plants, Cl toxicity is a much more important problem. Chloride ions have some roles in plant physiology: Photolysis of water at the oxidizing site of photosystem II, osmoregulation and stimulation of tonoplast proton-pumping ATPase, stomatal regulation, and N metabolism [Brown and Bassil, 2011], [Raven, 2017].

Plant species are divided into two in terms of Na tolerance: natrophilic and natrophobic [Smith et al., 2010a]. Natrophilic plants can behave as includers, accumulate and tolerate relatively high amounts of Na in their tissues and use it as an osmolyte whereas natrophobic plants cannot tolerate even modest increases in tissue Na levels. While Na may act as a beneficial element for plants at low concentrations, it is even considered an essential micronutrient for some C_4 plant species [Subbarao et al., 2003], [Brownell and Crossland, 1972]. In many Na-deficient C_4 plants, the flow of metabolites between mesophyll and bundle sheath cells is impaired [Brownell, 1980].

For many plants including crops like *Citrus* species, avocado, stone fruit trees, legumes, grapevine and cotton, Cl^- is the principle ion causing toxicity. Other crops including cereals like wheat and rice are mainly affected by Na^+ toxicity [Kronzucker et al., 2013]. Causes of ionic toxicity include disturbed hydration shells of macromolecules leading to structural and functional aberrations, impaired cellular ion and pH homeostasis, oxidative stress, and non-functional substitution of K^+ or

Ca^{2+} by Na^{+} [Smith et al., 2010a]. In addition to its direct adverse effects on plant growth as a toxic ion, a high soil Na^{+} concentration can also indirectly harm plants by degrading soil structure. Sodium-induced chemical soil compaction may deteriorate the aeration and water percolation properties of soils [Makoi and Verplancke, 2010].

Salinity causes nutritional disorders in plants through their effects on competitive uptake, nutrient availability, transport, and partitioning in plants [Hu and Schmidhalter, 2005]. As a result of the competition of toxic Na^{+} and Cl^{-} ions with ions such as K^{+}, Ca^{2+} and NO_3^{-}, mineral homeostasis is disrupted and symptoms of induced mineral deficiencies may be observed in plants. In plants exposed to salinity stress, N uptake and accumulation may be reduced as a result of the antagonistic interaction of Na^{+} with NH_4^{+} and Cl^{-} with NO_3^{-} [Bar et al., 1997]. Because of soil compaction, phosphorus availability is also reduced under saline conditions [Qadir and Schubert, 2002]. Potassium as a macronutrient is critical for protein synthesis, maintaining turgor pressure and providing resistance to salinity stress. However, because of the competition at uptake sites of roots, high Na concentration decreases the rate of K uptake and the K/Na ratio [Hu and Schmidhalter, 1997]. Therefore, the cellular and all plant-level functions of K are impaired. Also, tissue Ca levels are reduced in plants that suffer from salinity [Parihar et al., 2015], [Wei et al., 2003]. Therefore, the selectivity and integrity of plant cell membrane are disrupted and Na^{+} can passively enter the cell at an increased rate. Reduction of the tissue Ca/Na ratio under saline conditions inhibits plant growth and interferes with signal transduction events [Hussin et al., 2013]. Some micronutrient deficiencies are also observed in plants suffering from salinity [Rogers et al., 2003], [Hu and Schmidhalter, 2005]. The availability of most micronutrients decreases as a result of increased soil pH.

Reactive oxygen species (ROS) are produced as a by-product in the electron transfer chain reactions in mitochondria and phosynthetically active chloroplasts under normal conditions [Gill and Tuteja, 2010], [Bartosz, 1997]. Hydrogen peroxide (H_2O_2), despite being a ROS, is known to act as a signaling molecule between plant cells [Masood et al., 2012], [Mittler et al., 2004]. Excess ROS are eliminated by enzymatic and non-enzymatic antioxidants. However, these detoxification mechanisms in plants may prove insufficient under stress conditions, including salinity. Increased ROS levels cause an increase in the severity of primary (osmotic imbalance) and secondary (ion toxicity and ionic homeostasis) effects of salinity in

plants [Parida and Das, 2005], [Bohnert and Jensen, 1996]. Elevated ROS concentrations disrupt the integrity of cellular membranes. Therefore, it causes enhance lipid peroxidation, which is an indicator of oxidative damage [Menezes-Benavente et al., 2004]. The increased amount of ROS also increases protein degradation, membrane damage-induced cell death and disruption of photosynthetic enzymes and reduces stomatal conductance.

1.2.2. Plant Salinity Tolerance

The severity of the effects of salinity on plants varies depending on the light intensity, climatic condition, species, salt type, soil structure, genotype and stress exposure time [Tang et al., 2015]. Plants are divided into two according to their response to saline environments: glycophytes and halophytes. Regarding their salinity stress responses, they differ from each other in terms of toxic ion compartmentation and/or exclusion, osmotic adjustment, CO_2 assimilation, chlorophyll content, ROS production, antioxidant defense system, and hormonal regulation [Acosta-Motos et al., 2017].

Plants have developed 3 different mechanisms against salinity:

- Osmotic adjustment,
- Prevention of toxic ions from entering the vascular system (exclusion),
- Tolerating high tissue concentrations of potentially toxic ions (inclusion)

The osmotic effect of salinity causes a decrease in water uptake and thus the turgor pressure of plants. Osmotic adjustment is the accumulation of low molecular weight solutes and inorganic ions in cells in order to lower the solute potential and thus enable continued water uptake at low water availability [Turner et al., 2007]. Such adjustment provides sufficient turgor for growth and helps the plant resist salt stress and water deficit [Hare and Cress, 1997]. The organic low molecular weight solutes that contribute to osmotic adjustment are also called compatible solutes [Zhu, 2007]. Typical compatible solutes are proline, glycine betaine, glycerol, trehalose, pinitol, and mannitol [Jakobsen et al., 2007], [Chen and Jiang, 2010]. Compatible solutes may be neutral or zwitterionic and do not disrupt the hydration shells of biological molecules.

Osmotic adjustment is at least not the only beneficial effect of compatible solutes accumulation in higher plants [Ashraf and Foolad, 2007]. Further functions include antioxidative activity and protection of membrane and protein structures. For example, glycine betaine prevents the inactivation of Rubisco by toxic ions and stabilizes photosystem II. Trehalose stabilizes membranes and reduces the aggregation of denaturated proteins [López et al., 2008]. The accumulation of compatible solutes occurs in several ways: increased activity of biosynthetic enzymes, enhanced transport to tissues, increased availability of raw material, and reduced breakdown.

Plants can exclude toxic Na^+ ions from the cytosol thanks to Na^+ / H^+ antiport localized to the plasma membrane [Munns and Tester, 2008]. Exclusion of Na takes place in the roots, preventing translocation to the shoot of the plant [Acosta-Motos et al., 2017]. Most monocots and some halophytes are salt excluders. Saline vesicle glands in the epidermis allows the excretion of salts and prevents accumulation of toxic salt ions in the shoot of some species [Cheng and Hogarth, 2001]. In the long run, the exclusion strategy has some limitations:

- Secondary active antiporters exclude Na by consuming too much energy. In saline substrates, root cells have a higher carbohydrate and higher respiration rate requirement (energy cost) [Munns et al., 2006].
- Apoplast and rhizosphere become enriched with Na, which in turn aggravates the osmotic component salinity stress.

The third strategy for salinity resistance is ‘inclusion’. Plants that can implement this strategy tolerate high concentrations of Na^+ in tissue. Includers can exclude toxic Na^+ ions from the cytosol thanks to Na^+ / H^+ antiporters localized to the tonoplast of vacuole [Pardo et al., 2006].

Induction of an efficient antioxidative defense system also contributes to salinity tolerance [Zhu, 2007], [Gill and Tuteja, 2010]. Phytohormones including auxin, abscisic acid and gibberellin are also known to play critical roles in salinity responses [Egamberdieva, 2015], [Fahad et al., 2015].

1.3. Boron in Plants

Boron (B) is one of 14 elements that are essential for all vascular plants and is classified as a microelement in plant nutrition [Marschner, 2011]. Generally, the B requirement of dicots is higher than that of monocots. This difference is mostly due to the differences between the cell wall structures of cereals and dicotyledons. Boron primarily important for the formation, maintenance and function of the cell wall structure [Brown et al., 2002]. The cell wall is significantly affected in B-deficient plants, which are visible at macroscopic (cracked stem, stem corkiness) and microscopic scales (cell wall abnormalities) [Loomis and Durst, 1992], [Brown et al., 2002]. Boron does not intend to be involved directly in cell wall synthesis, but B may affect the incorporation of proteins, pectins, and/or precursors into the establishing and expanding cell wall. It stabilizes the pectin network to regulate cell wall pores. Rhamnogalacturonan II (RG-II) is a structurally complex pectic polysaccharide exist in the walls of developing plant cells [O'Neill et al., 1996], [Kobayashi et al., 1996]. Boron in the cell wall mainly cross-links the apiosyl residue in the A side chain of each of two neighboring monomeric RGII molecules to construct a dimeric B-dRGII pectin complex [Marschner, 2011]. Boron cross-linking with RG-II positively affects the physical and mechanical strength of the primary cell wall. Boron-deficient cell walls contain monomeric RG-II yet no cross-linked RG-II borate dimer [Fleischer et al., 1999]. Thus, B is vital for cell-to-wall adhesion and the organization of the cell's architectural integrity.

Boron is also responsible for ensuring the structural integrity of the plasma membrane and preventing potential damage to the membrane structures of ROS [Cakmak and Römheld, 1997]. Indirectly, B affects the amino acid metabolism, phloem transport of carbohydrates, photosynthesis, enzyme activation and auxin metabolism [Power and Woods, 1997]. Boron plays critical roles during seed germination, anther development, pollen tube formation and seed set [Wang et al., 2003].

Boron deficiency is considered to be the most widespread microelement deficiency problem in global crop production [Shorrocks et al., 1997]. However, B, as an essential micronutrient, has a very narrow adequacy zone; that is, the difference between B levels associated with B deficiency and those associated with B toxicity is

surprisingly small [Ben-Gal and Shani, 2002]. As B deficiency can be relatively easily corrected by careful soil and foliar fertilization practices, B toxicity is a harder-to-manage problem in crop production.

1.3.1. Boron Toxicity

Just like salinity stress, B toxicity observed in arid and semi-arid regions and may be exacerbated by certain anthropogenic activities such as using B-rich groundwater for irrigation [Kaur et al., 2006]. States reporting B toxicity in their soils are Turkey, Australia, US, Russia, Italy, most Middle East territories, some South East Asia countries like India, and some South American lands [Brdar-Jokanović, 2020]. Boron map of Turkey's soils created in 2010 from cooperation with the National Boron Institute (BOREN) and the Center for Soil Fertilizers and Water Resources Research Institute is updated in 2017 (Figure 1.2).

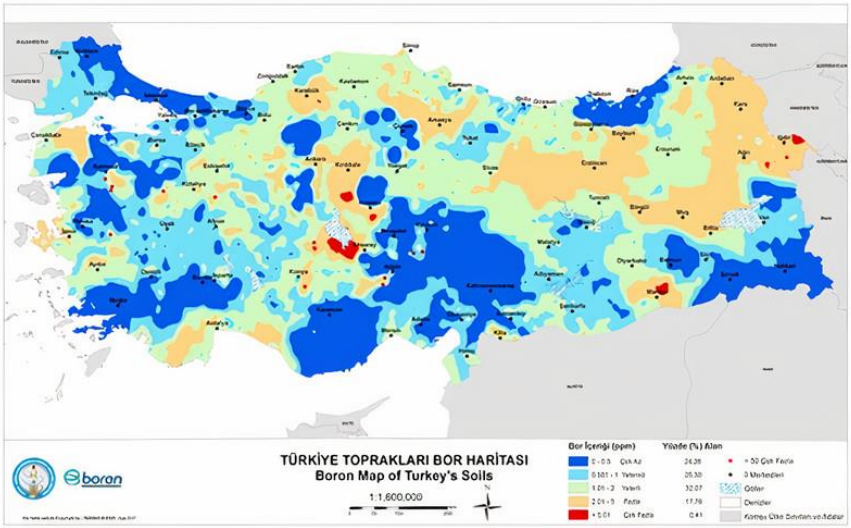

Figure 1.2: Boron Map of Turkey's Soils.

According to this map, soil B toxicity in Turkey has been determined in productive agricultural areas, especially in Central Anatolia. Indeed, soil B deficiency among agricultural areas in the World is more prevalent than soil B toxicity [Brdar-Jokanović, 2020]. Because of the extremely narrow B adequacy zone in plants, excessive B fertilization, which is used unconsciously to ameliorate B deficiency, usually causes B toxicity in plants.

Under real-life conditions, B toxicity occurs rarely as a stand-alone problem

but is rather often accompanied by salinity and drought [Grieve et al., 2010]. The susceptibility of crops to B toxicity depends on species and genotype [Edelstein et al., 2005]. Boron, which is carried from root to shoot by xylem, accumulates more in mature leaves where transpiration is more intense.

1.3.2. Boron Uptake and Distribution of Tissues

Boron is absorbed by plants through the roots as uncharged boric acid (H_3BO_3) [Raven, 1980]. Compared with all other elements, B is the only mineral nutrient taken up by roots as non-ionic form. Since B is a small, soluble, and uncharged molecule, it easily moves across the lipid bilayer and xylem [Brown and Shelp, 1997]. Boron absorption is closely linked to the external media B concentration. Under sufficient or excess B media, plants display a passive diffusion across lipid bilayers to B uptake into root cells to provide B requirement without energy consumption [Princi et al., 2015]. Even so, under restricted B availability, facilitated membrane transport and an energy-dependent high-affinity transport system are expected for B transport into the roots and toward xylem.

Boron absorption is highly influenced by soil water content and reduces in a dry environment where mass flow to the roots is diminished [Brdar-Jokanović, 2020]. In addition, the decreased B content in shoot due to the reduction of water potential and transpiration stream under saline conditions can be considered as evidence that B is transported by xylem [Princi et al., 2015]. Therefore, most of the B carrying with transpiration accumulates in the tips and edges of mature leaves.

After all, phloem also plays a role in supplying B to locations that do not lose water readily such as both shoot and generative tissues, depending on species [Brown and Shelp, 1997]. In many species including wheat and barley, B is immobile and cannot retranslocated to sink tissues. Boron has limited phloem mobility in plant species with sucrose as a primary photosynthates [Brdar-Jokanović, 2020]. In other species (apple, pear, sugar beet, etc.), it is quite mobile. The system of B transport via phloem occurred in the development of B-diol complex with sugar alcohols (sorbitol and/or mannitol), typically used for the phloem translocation of primary products of photosynthesis. Relatively higher B concentration in mature leaves suggests low B phloem mobility of the plant species. Otherwise, a higher B concentration in young leaves and fruits implies adequate B phloem mobility.

1.3.3. Effects of Boron Toxicity on Plants

Boron toxicity negatively affects many processes in plant cells [Reid et al., 2004]. Accumulated B in cytosol reacts with some important metabolites to form stable complexes and negatively affects plant growth and yield, causing metabolic deteriorations in cells. It affects the cell wall structure and disrupts integrity [Hajiboland et al., 2015]. Since B toxicity disrupts cell division, protein biosynthesis, integrity of lipids, and carbohydrate metabolism, it impairs the growth, development and energy mechanism of the cell [Sang et al., 2015], [Reid et al., 2004]. The chlorophyll content in the leaves decreases, stomatal conductance decreases, and the photosynthetic efficiency declines [Lovatt and Bates, 1984], [Ghanati et al., 2005]. Although it is assumed that B toxicity increases lipid peroxidation [Karabal et al., 2003], some studies reported that the activities of antioxidant enzymes are inconsistent under toxic-B conditions in different plant species and cultivars [Reid et al., 2004], [Cervilla et al., 2007].

Boron toxicity leads to chlorosis and necrosis in affected leaves, starting from tips and margins extending toward the leaf base. Impairment of shoot and root growth is observed. In addition, due to the effects of excess B on pollen development and pollen tube growth, seed and fruit set may be reduced, resulting in yield losses [Fang et al., 2016]. Whereas severe B toxicity may cause yield loss by reducing the photosynthetic area in wheat, slight B toxicity may not have a significant negative impact on yield [Brdar-Jokanović, 2020].

1.3.4. Plant Boron Toxicity Tolerance

Plant species and even cultivars within a species could vary in their B tolerance [Schnurbusch et al., 2010]. B tolerance may alter between plants and even cultivars because of the difference of critical toxicity concentration in leaves, phloem mobility, transpiration stream, internal mechanisms, and exclusion capabilities [Reid et al., 2004]. The process of B toxicity tolerance in plants involve B uptake from the soil, distribution within the shoot and generative tissues, accumulation at the end of transpiration flow, and heterogeneity within leaves.

It is one of the most important B tolerance mechanisms of the plants, that the B, which is adequate or excess in the media, is taken from the root by passive

diffusion and accumulates at the tips and edges of the old leaves by transpiration stream [Nable et al., 1997]. Therefore, necrotic and chlorotic symptoms are observed in tips and edges of old leaves where boron accumulates. This circumstance indicates that the plants able to self-sacrifice older leaves for younger leaves to continue their healthy growth. So, phloem immobility holds B away from vital metabolic sites, maintaining it instead in leaf tips and edges, where despite suffering necrosis, plants are still able to preserve the vast photosynthetic area [Nable et al., 1997].

However, B toxicity tolerance cannot be explained exclusively by transpiration, [Stangoulis and Reid, 2002]. Furthermore, it is known that there are internal tolerance mechanisms to reduce the severity of B toxicity in leaves. Internal strategies for ameliorating B toxicity include boron binding agents, B compartmentation in vacuoles, and B transport from sensitive symplastic compartments into the leaf apoplasm [Princi et al., 2015].

As mentioned before, under restricted B availability, facilitated membrane transport and an energy-dependent high-affinity transport system are expected for B transport into the roots. These energy-consuming transport systems also conduct both B uptake decline and excess B efflux to maintaining B tolerance [Brdar-Jokanović, 2020]. Shoot and root B concentration is lower in B-tolerant wheat cultivars than B-sensitive varieties because of the B efflux to soil and restricting the transfer of B to the shoot [Karabal et al., 2003], [Yau and Ryan, 2008]

For this reason, the breeding of varieties resistant to B toxicity and selecting resistant cultivars in areas affected by B toxicity are believed to be an effective way to tackle B toxicity in wheat production [Brown and Shelp, 1997], [Princi et al., 2015]. Also, avoiding the use of low-quality irrigation water high in B and excessive use of B-containing fertilizers are key strategies for the management of B toxicity.

1.4. Interaction Between Salinity and B toxicity

Salinity and B toxicity are frequently observed together in crops grown in arid and semi-arid regions due to limited leaching, especially in irrigated fields and orchards that depend on poor-quality irrigation water [Ben-Gal and Shani, 2002]. Salinity and B toxicity combination observed in plants is mainly due to irrigation with water containing high levels of salt and B or through the growth of plants in regularly salt and B-rich soils [Pandey et al., 2019]. This commonly occurring stress

combinations has been subject to research on many different crop species including wheat [Grieve and Poss, 2000], tomato and cucumber [Alpaslan and Gunes, 2001], and spinach and tomato [Gunes et al., 2007b]. In addition to inhibiting plant growth and causing yield loss by affecting various yield components, salinity and B toxicity impair germination parameters and limit root and shoot elongation while increasing membrane leakage [Ismail, 2004], [Molassiotis et al., 2006].

Due to the complex nature of the interactions between salinity and B toxicity, the effects of the combination of salinity and B toxicity on crops often differ from the simple sum of their individual effects. In the literature, there is an obvious inconsistency regarding the interactive effects of salinity stress and B toxicity on each other, which can aggravate, ameliorate or otherwise modulate each other's adverse effects.

In some studies, an antagonistic relationship was reported for salinity and B toxicity, implying that the two stresses could alleviate each other's adverse effects on plants when they co-occur. In a study conducted on corn (*Zea mays*) under hydroponic growth conditions, it was reported that aquaporins played an important role in B uptake under B toxicity conditions, and simultaneous salinity decreased the uptake of B by impairing the aquaporin function [Bastías et al., 2004]. As a result, in corn plants subjected to salinity and excess B conditions at the same time, tissue B accumulation was limited by salinity and the main cause for the observed negative effects on parameters such as growth and chlorophyll concentration was salinity.

Another study suggesting an antagonistic relationship between salinity and B toxicity was conducted on pepper (*Capsicum annuum* L.). In this study, B uptake decreased in the presence of toxic levels of Cl^- while Cl^- uptake decreased in the presence of toxic levels of B [Yermiyahu et al., 2008]. However, a physiological mechanism to explain such an antagonistic relationship between B and Cl uptake was not provided.

In a study of salinity x B toxicity interaction on broccoli (*Brassica oleracea* L.), toxic B levels were associated with reduced Na concentrations in leaves [Smith et al., 2010a]. Similarly, in an experiment with 4 different genotypes of bread wheat (*Triticum aestivum*), Naz et al. (2018) showed that the negative effect of salinity on wheat growth could be alleviated by increasing levels of B. Increased stress tolerance was attributed to enhanced antioxidative defense due to increased activities of enzymes including SOD and CAT and reduced toxic ion accumulation. It is

concluded that B fertilization in B-poor but saline soils may be utilized to ameliorate the negative effects of salinity.

In contrast to the studies mentioned above, it has also been claimed that the relationship between salinity and B toxicity in crop production is synergistic rather than antagonistic, implying that a co-occurrence of these stresses may in fact result in aggravated effects, which are worse than their separate effects. Combined salinity and B toxicity was found to increase the tissue accumulation of B in wheat compared to B toxicity alone [Wimmer et al., 2003]. Alpaslan and Gunes (2001) suggested a species-dependence of the effects of salinity X B toxicity interaction on B uptake and accumulation. It was observed that the uptake of B and the shoot B concentration in tomato were decreased by co-occurring salinity whereas higher shoot B concentrations were measured in cucumber subjected to salinity at the same time.

The effects of salinity on B accumulation in wheat (*Triticum aestivum*) were shown to be related to the level of B availability [Masood et al., 2012]. Salinity increased the tissue B concentrations under adequate-B conditions while it decreased them under toxic-B conditions.

Yet other researchers suggested that salinity and B toxicity affect crops independently even when they co-occur. In a study conducted at various B and salinity levels, no clear interaction was found between salinity and B toxicity in wheat [Bingham et al., 1987]. Effects of salinity and excess B on melon were also reported to be largely independent from each other [Edelstein et al., 2005].

1.5. Wheat

Wheat is among the most important crops for global food supply. In developing countries, including Turkey, wheat alone provides 40% of daily calorie needs [Cakmak, 2008]. However, wheat is often grown in stress-prone and marginal environments where higher-value crops cannot be grown successfully. Therefore, as a result of various abiotic stresses and their combinations, wheat growers suffer from great yield losses.

The most cultivated wheat species in the world are bread (common) wheat (*Triticum aestivum*, 2n=6X=42) and durum wheat (*Triticum durum*, 2n=4X=28). These two wheat species, which even have separate chromosome numbers, naturally differ in terms of stress resistance, yield, and development [Marti and Slafer, 2014].

It is known that when the EC_e rises above 4.5 $dS.m^{-1}$ salinity starts to adversely affect the dry weight (DW) of bread wheat because of the reach salt tolerance line (threshold) and salinity will decrease DW by 2.6 % per 1 unit $dS.m^{-1}$ increase (slope) [Maas and Grattan, 2015], [Francois et al., 1986]. However, the durum wheat DW threshold value is assumed that 2.1 $dS.m^{-1}$ and the slope is 2.5 %. The effect of salinity on the vegetative growth of wheat is harsher than its effect on generative development [Marti and Slafer, 2014]. If the soil's ECe value is 8.6, salinity could decline the bread wheat grain yield, although this value is 5.9 for durum wheat [Maas and Grattan, 2015], [Francois et al., 1986]. Bread and durum wheat slope values are 3.0 and 3.8, respectively.

Considering the differences in cultivars and developmental stages in wheat, it is ideal that the shoot B concentration is in the range of 5-15 $mg.kg^{-1}$ [Marschner, 2011]. We should worry about toxicity when the B measured in the wheat reaches 15 $mg.kg^{-1}$ in shoot and 10-40 $mg.kg^{-1}$ in grain. In fact, B concentration may achieve 1500 $mg.kg^{-1}$ in the necrotic tips and edges of the leaves under toxic-B conditions [Nable et al., 1997]. Also, in literature, although cultivars show little difference, durum wheat species were found to be more sensitive to B toxicity than most bread wheat cultivars [Kalayci et al., 1998]. In addition, it should be noted that phloem mobility of B is very low in wheat [Brown et al., 2002].

Bread wheat tended to be more efficient than durum wheat under stressful circumstances and vice versa durum wheat to be an alternative to bread wheat in high-yielding conditions when fertilized sufficiently in non-stressed conditions to meet the requirements of protein concentration from the industry [Marti and Slafer, 2014].

It is known that the salinity and B toxicity stress factors mentioned above are highly effective on both wheat species and cause physiological and nutritional changes as a result of the interaction of these two agents [Pandey et al., 2019].

2. MATERIALS AND METHODS

2.1. Materials and General Information

2.1.1. Plant Material

In experiments documented here, Nusrat, a Turkish bread wheat cultivar (*Triticum aestivum* cv. Nusrat), Adana-99, a Turkish bread wheat cultivar (*Triticum aestivum* cv. Adana-99), Saricanak, a Turkish durum wheat cultivar (*Triticum durum* cv. Saricanak), and Fuatbey-2000, a Turkish durum wheat cultivar (*Triticum durum* cv. Fuatbey-2000) were cultivated as described below.

All wheat cultivars were selected in spring character and can be well adapted to hydroponic and soil experiments conducted under growth chamber and greenhouse conditions [Kalayci et al., 1998], [Marti and Slafer, 2014]. Nusrat is a bread wheat cultivar with great yield potential and it was verified in 2015 and it is expected that Nusrat cultivation will become common in the Turkey [Web 1, 2021]. The grains of Adana-99, the other bread wheat variety, are coated with zinc (Zn). Adana-99 is known to be very resistant to drought and cold, and its average yield is 665 $kg.da^{-1}$ [Web 2, 2021]. Saricanak durum wheat variety was verified approximately 25 years ago and is widely preferred by farmers in the southeastern Anatolia region in Turkey [Web 3, 2021]. Fuatbey-2000, another durum wheat variety, is also grown in similar regions and is a highly productive variety [Web 4, 2021].

2.1.2. Sodium Chloride (NaCl)

Saline conditions were created by adding sodium chloride (NaCl) to the growth medium in all experiments. Different EC increments were obtained by using different concentrations of NaCl concentrations in addition to mineral fertilizers.

2.1.3. Boron (B) Toxicity

In both soil and solution culture experiments, B toxicity was studied by adding excess B in the form of boric acid (H_3BO_3) to a B-sufficient control medium.

2.2. Plant Growth Facilities

2.2.1. Greenhouse

Soil culture experiments were conducted in a greenhouse under natural daylight. The geographic coordinates of the greenhouse are 40º 53' 24.5" N and 29º 22' 46.7" E. The greenhouse is equipped with a heating system and an evaporative cooling system, which keeps the temperature inside the greenhouse in the range of 15-25°C depending on the season and daytime.

2.2.2. Growth Chamber

The solution culture experiments, and some soil culture experiments were conducted in a growth chamber under controlled climatic conditions (light/dark periods: 16/8 h; temperature (light/dark): 25°C/20°C; relative humidity (light/dark): 60%/70%).

2.3. Plant Growth Cultures

2.3.1. Solution Culture

Seeds were germinated in perlite moisturized with 4 mM $Ca(NO_3)_2.4H_2O$ solution for 5-6 days under growth chamber conditions and then, seedlings were transferred to 4.5 L black plastic hydroponics pots. The full strength nutrient solution consisted of 0.6 mM K_2SO_4, 4 mM $Ca(NO_3)_2.4H_2O$, 0.2 mM KH_2PO_4, 0.75 mM $MgSO_4.7H_2O$, 0.1 mM KCl, 75 µM Fe (in the form of FeEDTA), 2 µM H_3BO_3, 2 µM $MnSO_4.H_2O$, 3 µM $ZnSO_4.7H_2O$, 0.6 µM $CuSO_4.5H_20$, 0.5 µM $(NH_4)6Mo_7O_{24}.4H_2O$. All the chemicals used for his purpose were of analytical grade. For the first 3 days after transfer to hydroponics, half-strength solution was used. Then, the solutions were replaced with full-strength nutrient solution. Nutrient solutions were continuously aerated and refreshed every 7 days [Kutman, 2010].

2.3.2. Soil Culture

In the preliminary soil experiment conducted under growth chamber conditions, seeds were sown in plastic pots each containing 1.25 kg air-dry soil whereas in other soil experiments conducted in glasshouse, seeds were sown in larger plastic pots, each filled with 3.5 kg air-dry soil. The experimental soil was transported from Tepeören, Tuzla, Istanbul. This soil has a clay-loam structure, low organic matter content and a slightly alkaline pH (7.5 in 1:5 soil-water extract). The electrical conductivity of the saturated paste extract of unfertilized soil (EC_e) is estimated to be 1 dS/m.

Before sowing the seeds, pots in all treatment groups were fertilized with 100 ppm P (in the form of KH_2PO_4) and 250 ppm N (in the form of $Ca(NO_3)_2$). Basal fertilizers were added as concentrated solutions and incorporated into soil by vigorous mixing. In experiments in which plants were to be grown to full maturity for harvesting grains, 150 mg/kg N was applied to the soil in the form of calcium nitrate $Ca(NO_3)_2$ together with irrigation water during anthesis.

Pots were irrigated with dH_2O regularly until the experiments were finished. Experiments had full factorial designs with 4 independent pot replicates for each treatment and the replicates were randomized in the greenhouse to account for the possibility of local variations in conditions. In each pot, 15 wheat seeds were sown, and the seedlings were thinned to 5 or 8 per pot, depending on the experiment, shortly after emergence.

2.4. Sampling and Harvest

Fresh samples were taken from plants with cutting desired leaves. These fresh samples were used in REL analysis described below.

In addition, for obtain dry sample, shoot and root of wheat were harvested and dried at 65°C. Dried and finely ground plant samples were used for element analysis and glycine betaine determination.

Moreover, at the end of the experiment, second and third youngest leaves were cut and pooled. From each pot replicate, 0.5 grams of fresh samples were taken and after shocking by using liquid nitrogen the samples were stored at -80°C until they

were used for extraction. Frozen plant samples were used for spectrophotometric assays except glycine betaine method.

All harvested plant samples except spikes were washed in dH_2O and dried at 65°C at the end of third (yield) soil experiment. Threshing of grains was done by laboratory-size thresher. Grains were dried at 45°C. Dried samples were weighed at room temperature for biomass and yield determination. Also, dried straw and grain samples were used for determine element analysis [Kutman, 2010].

2.5. Experimental Designs and Applied Methods

2.5.1. Solution Culture Experiments

2.5.1.1. First Solution Culture Experiment

In this experiment, bread wheat (*Triticum aestivum* cv. Nusrat) plants were grown in solution culture under controlled climatic conditions. The solution culture system and the climatic conditions in the growth chamber were described in '2.3.1' and '2.2.1.', respectively. For each treatment, there were 4 pot replicates and 12 single plants in each pot. The experiment was designed as a total of 48 pots (4 replicate x 4 salinity levels x 3 B toxicity levels). Plants were grown at four salinity levels: 0, 40, 80, and 120 mM salt in the form of NaCl. Also, plants were grown at three B toxicity levels: Adequate B pots were supplied with 0 mM B, moderate B pots with 0.5 mM B, and severe B pots with 2.5 mM B in the form of H_3BO_3. The first salinity and B toxicity treatments were utilized 14 days after sowing (DAS). Afterward, the second stress treatments were applied 21 DAS.

The experiment was terminated 28 DAS. The shoot and root of each pot were harvested separately. Harvested roots were washed with deionized water. All samples were dried, weighed, and analyzed for mineral concentrations as described in below.

2.5.1.2.Second Solution Culture Experiment

In this experiment, bread wheat (*Triticum aestivum* cv. Nusrat) plants were grown in solution culture under controlled climatic conditions. The solution culture system and the climatic conditions in the growth chamber were described in above. For each treatment, there were 4 pot replicates and 12 single plants in each pot. The experiment was designed as a total of 16 pots (4 replicate x 2 salinity levels x 2 B toxicity levels). Plants were grown at two salinity levels: Low salt pots were supplied with 0 mM NaCl, and high salt pots with 80 mM NaCl. Also, plants were grown at two B toxicity levels: 0, and 1 mM B in the form of H_3BO_3. The first salinity and B toxicity treatments were applied 14 days after sowing (DAS). Then, the second stress treatments were done 21 DAS.

Necrosis severity at the tip of each leaf was measured before harvest. The experiment was completed 28 DAS. The shoot of each pot was harvested. The leaves in each pot were divided into 3 parts: Young leaves, necrotic parts of older leaves, and rest of older leaves. All samples were dried, weighed and analyzed for mineral concentrations as described in below.

2.5.1.3.Third Solution Culture Experiment

In this experiment, bread wheat (*Triticum aestivum* cv. Nusrat) and durum wheat (*Triticum durum* cv. Saricanak) plants grown in solution culture under controlled climatic conditions. The solution culture system and the climatic conditions in the growth chamber were described in '2.3.1' and '2.2.2.', respectively. For each treatment, there were 4 pot replicates and 15 single plants in each pot. The experiment was designed as a total of 48 pots (4 replicate x 3 salinity levels x 2 B toxicity levels x 2 plant species). Plants were grown at three salinity levels: Low salt pots were supplied with 0 mM NaCl, medium salt pots with 60 mM NaCl, and high salt pots with 120 mM NaCl. Also, plants were grown at two B toxicity levels: 0, and 1 mM B in the form of H_3BO_3. The first salinity and B toxicity treatments were done 14 DAS. The second time stress treatments were applied 21 DAS. The third stress treatments were utilized 28 DAS.

During the experiment, the rate of transpiration was measured 4 times as described in below. The first of these measurements was made 1 day before the stress

was applied, that is, 6 days after the transfer of plants to the solution culture. The second measurement was made immediately after the stress application, the third measurement was made one week after the stress application and the last one was the last day before harvest. Necrosis severity at the tip of each leaf was measured before harvest. The oldest second fresh leaf of one of the fifteen plants was used for the REL analysis at harvest day by the method described in below.

The experiment was completed 30 DAS. The shoot and root of each pot were harvested separately. Harvested roots were washed with deionized water. 12 plant samples of each pot were dried, weighed and analyzed for mineral concentrations as described in below. Also, shoot samples that were dried and ground into a fine powder were used for the determination of glycine betaine (GB) concentration as described in below.

The 3 seedlings of each pot were used as frozen sample as described above. After the homogenization, extracts were used for the determination of antioxidative enzyme activities, protein concentration, free proline concentration, and malondialdehyde (MDA) concentration as described below.

2.5.2. Soil Culture Experiments

2.5.2.1.First Soil Culture Experiment

In this experiment, bread wheat (*Triticum aestivum* cv. Nusrat) plants grown in soil culture under controlled climatic conditions. For each treatment, there were 4 pot replicates and 15 single plants in each pot. The experiment was designed as a total of 48 pots (4 replicate x 3 salinity levels x 4 B toxicity levels).

Plants were grown at three soil salinity levels throughout the development: 3.3 (control), 5.8, and 8.3 $dS.m^{-1}$ EC_e. It was calculated that 0.375 g of salt was required to increase the EC_e of 1 kilogram of soil by 1 ds/m. The amount of salt required to increase 2.5 ds/m for 1.25 kilograms of soil was determined. Also, plants were grown at four B toxicity levels: 0, 10, 20, and 30 $mg.kg^{-1}$ B in the form of H_3BO_3. Before sowing the seeds, stresses and fertilizers were mixed with the soil.

After 25 days of growth, shoots of each pot were harvested. All samples were dried, weighed and analyzed for mineral concentrations as described in below.

2.5.2.2. Second Soil Culture Experiment

In this experiment, bread wheat (*Triticum aestivum* cv. Nusrat and Adana-99) and durum wheat (*Triticum durum* cv. Saricanak and Fuatbey-2000) plants grown in soil culture under greenhouse conditions. The soil was prepared as described in above. For each treatment, there were 4 pot replicates and 15 single plants in each pot. The seedlings were thinned to 8 per pot, shortly after emergence. The experiment was designed as a total of 144 pots (4 replicate x 4 cultivar x 3 salinity levels x 3 B toxicity levels).

Plants were grown at three soil salinity levels throughout the development: 3.5 (control), 6, and 8.5 $dS.m^{-1}$ EC_e. It was calculated that 0.375 g of salt was required to increase the EC_e of 1 kilogram of soil by 1 ds/m. The amount of salt required to increase 2.5 ds/m for 3.5 kilograms of soil was determined. Also, plants were grown at three B toxicity levels: 0, 15, and 30 $mg.kg^{-1}$ B in the form of H_3BO_3. Before sowing the seeds, stresses and fertilizers were mixed with the soil.

After 40 DAS, shoots of each pot were harvested. Four plant samples of each pot were dried, weighed and analyzed for mineral concentrations as described in below. Also, shoot samples that were dried and ground into a fine powder were used for the determination of GB concentration as described in below.

The four seedlings of each pot were used as frozen sample. After the homogenization, extracts were used for the determination of antioxidative enzyme activities, protein concentration, free proline concentration, and MDA concentration as described below.

2.5.2.3. Third Soil Culture Experiment

In this experiment, bread wheat (*Triticum aestivum* cv. Nusrat and Adana-99) and durum wheat (*Triticum durum* cv. Saricanak and Fuatbey-2000) plants grown in soil culture under greenhouse conditions. The soil was prepared as described in 'Soil Culture'. For each treatment, there were 4 pot replicates and 15 single plants in each pot. The seedlings were thinned to 5 per pot, shortly after emergence. The experiment was designed as a total of 144 pots (4 replicate x 4 cultivar x 3 salinity levels x 3 B toxicity levels).

Plants were grown at three soil salinity levels throughout the development: 3.5 (control), 5.5, and 7.5 $dS.m^{-1}$ EC_e. It was calculated that 0.375 g of salt was required to increase the EC_e of 1 kilogram of soil by 1 ds/m. The amount of salt required to increase 2 ds/m for 3.5 kilograms of soil was determined. Also, plants were grown at four B toxicity levels: 0, 10, and 20 $mg.kg^{-1}$ B in the form of H_3BO_3. Before sowing the seeds, stresses and fertilizers were mixed with the soil.

When the plants senesced fully and the grains reached full maturity, the spikes and the straws were harvested separately described above. Threshing of grains was done by laboratory-size thresher. Straw and grain were dried, weighed and analyzed for mineral concentrations as described in below.

2.6. Element Analysis

Dried shoot and straw samples were finely ground with an electric coffee grinder. Also, grain samples of wheat analyzed but they don't grinded. Approximately 0.2 g of the dried and ground plant samples were weighed and placed in microwave digestion tubes. On top of each sample, 2 ml of 30% hydrogen peroxide (H_2O_2) and 5.0 ml of nitric acid (HNO_3) were added, and the digestion was conducted in a closed-vessel microwave system (MarsExpress; CEM Corp., Matthews, NC, USA). After cooling down sufficiently, total sample volume was finalized to 20 ml by adding double-deionized water and samples were filtered through analytical filter papers (Macherey-Nagel, Ø125 mm, blue band). Inductively coupled plasma optical emission spectrometry (ICP-OES) (Vista-Pro Axial, Varian, Australia) was used to determine the concentrations of macro- (except N) and micronutrients in digested plant samples. The accuracy of element analyses was checked by using certified standard reference materials obtained from the National Institute of Standards and Technology (Gaithersburg, MD, USA) [Kutman, 2010].

2.7. Gravimetric Determination of Transpiration Rate

The transpiration rate of hydroponically grown plants was determined gravimetrically. 24 hours before starting with data collection, at about 10:00 am, nutrient solutions in pots were refreshed such that the net weight of nutrient solution in each pot was exactly 4500 g. To determine the transpiration during the day, the net

weight of nutrient solution in each pot was measured 6 h later, the amount of water lost by transpiration in 6 h was calculated as the difference from the initial value (4500 g) and divided by 6 to calculate the transpiration rate per h. Another weight measurement was taken for each pot at exactly 24 h to also account for the rest of the day, including the night, and calculate the transpiration rate per day.

2.8. Determination of Relative Electrolyte Leakage (REL)

Fresh leaf samples were firstly washed with dH_2O and then placed in centrifuge tubes containing 50 ml of dH_2O so that the whole leaves remained submerged in water. After the tubes containing the leaves and water were kept at 25°C for 6 hours, the EC of the water was measured by an EC meter and the EC_1 value was determined for each sample. Then, the tubes with their caps tightly closed were kept at in a water bath set to 100°C for 30 minutes and the EC_2 value was measured for each sample after the tubes reached room temperature. Finally, the REL value was calculated as follows:

$REL = (EC_1/EC_2)*100\%$.

2.9. Spectrophotometric Assays

Antioxidative enzyme activities, total protein, lipid peroxidation, glycine betaine, and proline concentrations were determined with spectrophotometric protocols described below. The measurements were performed by using a UV-Visible spectrophotometer (Cary 300 Bio; Varian, Australia). Frozen fresh leaf samples were homogenized with the help of a ball mill (MM200; Retsch, Haan, Germany) by using liquid nitrogen and extraction buffer.

2.9.1. Antioxidative Enzymes and Total Protein Analysis

0.5 g frozen leaf samples were homogenized with liquid nitrogen and 50 mM potassium phosphate buffer (K-P) which described below. By mixing 50 mM KH_2PO_4 and 50 mM K_2HPO_4 in a 2:1 volume ratio, K-P buffer with a pH of 7.6 was prepared. The extraction buffer was prepared by adding 0.1 mM Titriplex III to this

K-P buffer. After the homogenization, the homogenates were centrifuged at 4600 min^{-1} for 15 min at 4°C, and the supernatants were transferred to microcentrifuge tubes, which were centrifuged again at 15.000 min^{-1} for 15 min at 4°C. These supernatants were used for antioxidative enzyme (superoxide dismutase (SOD), glutathione reductase (GR), ascorbate peroxidase (APX), catalase (CAT)) and Bradford protein assays.

2.9.1.1. Superoxide Dismutase (SOD) Analysis

Superoxide dismutase activity was determined following the method [Giannopolitis and Ries, 1977] 2.9 ml of 50 mM K-P extraction buffer was mixed with 0.5 ml of 50 mM Na_2CO_3, 0.5 ml of 13 mM L-methionine, 0.5 ml of 63 µM NBT, 0.1 ml of crude sample extract (1:10 diluted) and 0.5 ml of 1.3 µM riboflavin in glass tubes, respectively. The chemicals used were kept in the dark during preparation because they are sensitive to light. After riboflavin addition, glass tubes were placed in the growth chambers and kept under light for 8 min. The measurements were performed at 560 nm wavelength by using a sipper.

2.9.1.2. Glutathione Reductase (GR) Analysis

Glutathione reductase activity was determined following the method [Carlberg and Mannervik, 1985]. To determine the activity of the GR enzyme, 0.7 ml of 50 mM K-P buffer, 0.1 ml of 0.5 mM Oxidized Glutathion (GSSG), and 0.1 ml of crude sample extract (1:10 diluted) were mixed and finally 0.1 ml of 0.45 mM NADPH-Na_4 was added to this mixture. Mixture was transfer to the quartz cuvette. The activity was determined spectrophotometrically by using 340 nm wavelength and absorbance values were monitored for 2 min to find the average depletion rate of NADPH-Na_4.

2.9.1.3. Ascorbate Peroxidase (APX) Analysis

Ascorbate peroxidase activity was determined following the method [Nakano and Asada, 1981]. Assay medium included 0.7 ml of 50 mM K-P buffer was mixed with 0.1 ml of 12 mM H_2O_2 and 0.1 ml of crude sample extract (1:40 diluted), and

then 0.1 ml of mM ascorbic acid ($C_6H_8O_6$) in quartz cuvette. To calculate the average depletion rate of L-ascorbic acid, absorbance values at 290 nm were measured for 2 min with the spectrophotometer.

2.9.1.4. Catalase (CAT) Analysis

Catalase activity in shoot tissues of plants were determined according to protocole [Chance and Maehly, 1955] with slightly modification: H_2O_2 is used as 100 mM. 0.8 ml of 50 mM K-P buffer was mixed with 0.1 ml of 100 mM H_2O_2 and 0.1 ml of crude sample extract (1:80 diluted). The change in the absorbance of this mixture in quartz cuvette was followed for 2 min at 240 nm to calculate the average rate of H_2O_2 breakdown.

2.9.1.5. Measurement of Total Protein Concentration

Total protein concentration was identified following the method [Bradford, 1976]. To prepare bradford reagent, 0.1 g coomassie brilliant blue G-250 was dissolved in 50 ml ethanol and was mixed with 100 ml 85% ortho-phosphoric acid. This mixture was filtered and after filtration 100 ml glycerin was added to the reagent and the volume was brought to 1000 ml with dH_2O. The reagent was kept at 4°C for 24 h and then used for the assay. Protein standards (0, 100, 200, 400 and 800 ppm) were prepared by dissolving bovine serum albumin in K-P buffer. 5 ml of reagent was added to 0.1 ml sample (1:10 diluted) or standard and vortexed. After 10 minutes, the reading of the protein concentration was performed at 595 nm by sipper for standards and samples.

2.9.2. Determination of Malondialdehyde (MDA) Concentration

Lipid peroxidation was determined by measuring malondialdehyde (MDA) concentration [Hodges et al., 1999]. The extraction buffer for MDA analysis was prepared (80% ethanol:dH_2O). 0.5 g of frozen leaf samples were homogenized with the help of a ball mill by using liquid nitrogen and 12.5 ml of 80% ethanol. The homogenates were centrifuged at 3000 g for 10 min at 4°C, and the supernatants

were transferred to microcentrifuge tubes. These supernatants were used for the determination of MDA concentrations.

After the preparation of crude plant extracts for MDA assay, -TBA and +TBA solutions were prepared separately. -TBA solution include: 0.5 mM butylated hydroxytoluene (BHT) and 20% trichloroacetic acid (TCA). +TBA solution include: 0.5 mM BHT, 20% TCA, and 0.65% thiobarbituric acid (TBA). 1 ml of - and + TBA were transferred to 2 ml microcentrifuge tubes with screw cap separately for each sample. 1 ml aliquot of sample was added to each of the 2 microcentrifuge tubes (for - and + TBA). It is mixed by inversion or vortexing. Microcentrifuge tubes were placed in racks and kept in a hot water bath at 95°C for 25 minutes. Microcentrifuge tubes were centrifuged at 3000 min^{-1} for 10 minutes at 4° C. The absorbance of supernatants in in quartz cuvette was read at 440, 532 and 600 nm by spectrophotometer for determining MDA concentration. Malondialdehyde concentration were calculated with an equation.

2.9.3. Determination of Free Proline Concentration

Proline concentration was measured by ninhydrin method [Bates et al., 1973]. The extraction buffer for free proline analysis was prepared (3% sulfosalicylic acid:dH_2O). 0.5 grams of frozen fresh leaf samples were homogenized with the help of a ball mill by using liquid nitrogen and 10 ml of 3% sulfosalicylic acid buffer. The homogenates were centrifuged at 5000 g for 15 min at 4°C, and the supernatants were transferred to 2 ml e microcentrifuge tubes.

2 ml of the aliquot of sample were mixed into a tube with 2 ml of glacial acetic acid and 2 ml of acid-ninhydrin solution (1.25 g of ninhydrin was dissolved by heating in 30 ml of glacial acetic acid and 20 ml of 6 M phosphoric acid). After incubating at 100°C for 1 hour, it was cooled by standing in an ice bath. After the mixture had cooled, 4 ml of toluene was added, and this new mixture was vortexed. The toluene phase containing the chromophore was used for free proline analysis. Free proline standards (0, 10, 20, 40 and 50 ppm) were prepared by dissolving proline in 3% sulfosalicylic acid. The absorbance values of the standards and samples in quartz cuvette at 520 nm measured and the concentration of free proline in the samples determined by drawing the standard curve.

2.9.4. Determination of Glycine Betaine (GB) Concentration

Glycine betaine concentration in the shoot samples were determined by using the assay described by Grieve and Grattan, 1983. 20 ml of dH_2O was added to 0.5 g dried and finely ground shoot sample. Using an orbital shaker at 300 rpm for 24 hours at room temperature, water-soluble components were extracted. The mixture was centrifuged at 5000 min^{-1} for 10 minutes at room temperature. 1 ml of 2 N sulfuric acid was added to 1 ml of supernatant. A 0.5 ml aliquot from this mixture was cooled in an ice bath for 1 hour. Also, cold KI-I_2 reagent, prepared by dissolving 15.7 g of iodine and 20.0 g of KI in 100 ml dH_2O was added and the reactants were gently stirred with a vortex. 0.2 ml of cold KI-I_2 reagent was added on the ice-cold aliquot of shoot. The mixture was kept at 4°C for 16 hours and then centrifuged at 4000 min^{-1} for 15 minutes at 0 °C. The liquid phase was carefully removed from the medium and the per-iodide crystals at the bottom were dissolved by adding 9 ml of 1,2-dichloroethane and vortexing the tubes. Glycine betaine standards (0, 50, 100, 200 and, 400 ppm) were prepared and processed like samples. The absorbance of the samples and standards in quartz cuvette at 360 nm was measured using spectrophotometer. Glycine betaine concentrations in the samples were calculated using the standard curve drawn.

2.10. Calculations

The mineral concentrations other than N in the samples were calculated by multiplying the values measured by ICP-OES with the dilution factor, which is calculated for each sample separately by dividing the total sample volume (ml) by the dry weight (g) of the digested sample.

For calculating the mineral contents for a given plant part, the calculated mineral concentrations were multiplied by the measured total dry weights of the concerned plant part. Similarly, the grain mineral yields, i.e. the total amounts of minerals of interest deposited in the grains, were determined by multiplying the grain yield by the grain mineral concentrations.

The harvest index (%) was calculated by dividing the grain yield by the sum of the grain yield and dry straw biomass.

Shoot water content (WC) was calculated in accordance with the formula as follows: Water content (WC) = (Fresh weight – Dry weight)/Dry weight.

2.11. Statistical Analysis

All experiments had factorial designs and 4 replicates in each treatment group. For statistical analysis, the JMP software was used. The significance of the effects of the treatments and their interactions on the reported traits was evaluated by analysis of variance (ANOVA) at 5 % significance. Significant differences between means were then determined using Tukey's honestly significant difference (HSD) test at 5 % significance.

3. RESULTS

3.1. Hydroponic Experiment Results

3.1.1. First Hydroponic Experiment Results

The first hydroponic experiment was conducted to study the effects of the interaction of different levels of salinity stress with different levels of B toxicity on growth, leaf symptoms and shoot and root accumulation of minerals in bread wheat under solution culture conditions. Another aim of the experiment was to determine salinity and B toxicity levels which would allow an in-depth physiological investigation of the physiological aspects underlying the effects of this interaction on wheat in subsequent experiments under hydroponic conditions.

Just one week after the start of stress application, severe B toxicity caused chlorosis and necrosis in mature leaves of experimental plants beginning from leaf tips and margins. This symptom was alleviated by increasing levels of salinity. However, increasing salinity also decreased plant growth under severe B toxicity (Figure 3.1).

Figure 3.1: Effects of severe B toxicity supply (2.5 mM) on 3-weeks-old bread wheat (*Triticum aestivum* cv. Nusrat) grown hydroponically at different levels of salinity (no: 0 mM NaCl; low: 40 mM NaCl; medium: 80 mM NaCl; high: 120 mM NaCl) under growth chamber conditions.

Especially in the absence of salinity, it was observed that two weeks after the start of the stress treatments, necrosis progressed dramatically under severe B toxicity and caused the tips of affected leaves to hang down. In contrast, it was realized that increasing B toxicity under the high salinity condition gave plants a stiffer appearance. Also, marked reduction in root growth was observed with increasing level of B toxicity in the absence of salinity.

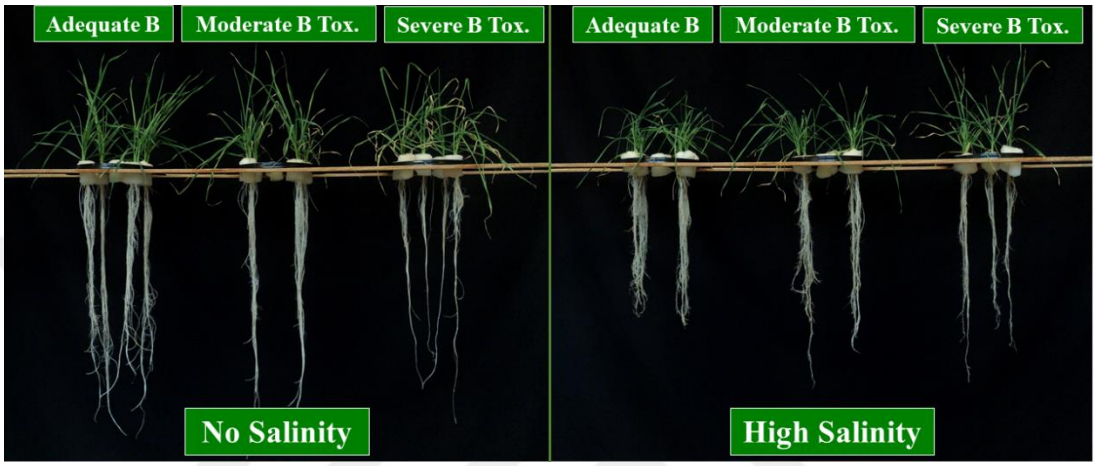

Figure 3.2: Shoot and root growth and general appearance of 4-weeks-old bread wheat (*Triticum aestivum* cv. Nusrat) grown hydroponically at different levels of B supply (adequate: 2 µM; moderate toxicity: 0.5 mM; severe toxicity: 2.5 mM), depending on absence or presence of salinity stress (no: 0 mM NaCl; high: 120 mM NaCl).

Table 3.1: Analysis of variance of the main effects of salinity and B supply and their interaction on dry weights (DW), selected mineral concentrations and contents and K/Na ratios of shoots and roots of 4-weeks-old bread wheat (*Triticum aestivum* cv. Nusrat) grown hydroponically under growth chamber conditions.

Source of Variation	**Shoot DW**	**Root DW**	**Shoot K/Na Ratio**	**Root K/Na Ratio**
Salinity (S)	**	*n.s.*	***	***
B Supply (BS)	*n.s.*	***	*n.s.*	*n.s.*
S*BS	*n.s.*	*n.s.*	*n.s.*	*n.s.*
Source of Variation	**Shoot B Conc.**	**Shoot B Content**	**Root B Conc.**	**Root B Content**
Salinity (S)	***	***	*n.s.*	*n.s.*
B Supply (BS)	***	***	***	***
S*BS	***	***	**	**
Source of Variation	**Shoot Na Conc.**	**Shoot Na Content**	**Root Na Conc.**	**Root Na Content**
Salinity (S)	***	***	***	***
B Supply (BS)	*	**	*n.s.*	*n.s.*
S*BS	*n.s.*	*	*n.s.*	*n.s.*
Source of Variation	**Shoot K Conc.**	**Shoot K Content**	**Root K Conc.**	**Root K Content**
Salinity (S)	**	***	*n.s.*	*n.s.*
B Supply (BS)	*	*n.s.*	*n.s.*	*
S*BS	*n.s.*	*n.s.*	*	*n.s.*
Source of Variation	**Shoot Ca Conc.**	**Root Ca Conc.**	**Shoot Mg Conc.**	**Root Mg Conc.**
Salinity (S)	***	***	***	**
B Supply (BS)	***	*n.s.*	***	**
S*BS	*n.s.*	*n.s.*	*n.s.*	*n.s.*

n.s. Not significant; * $0.01 \leq F\ Pr. < 0.05$; ** $0.001 \leq F\ Pr. < 0.01$; *** $F\ Pr. < 0.001$

Shoot dry weight (DW) was decreased significantly by increasing salinity stress while B toxicity had no significant effect on shoot dry weight (Table 3.1; Table 3.2). In contrast, root DW was not affected by salinity stress but it was reduced significantly by increasing B toxicity.

Table 3.2: Effects of different levels of salinity (none: 0 mM NaCl; low: 40 mM NaCl; medium: 80 mM NaCl; high: 120 mM NaCl) and B supply (adequate: 2 μM; moderate toxicity: 0.5 mM; severe toxicity: 2.5 mM) on shoot and root dry weights (DW) of 4-weeks-old bread wheat (*Triticum aestivum* cv. Nusrat) grown hydroponically under growth chamber conditions. Data points represents the means and standard deviations of four replicates.

Salinity	**B Supply**	**Shoot DW ($mg.plant^{-1}$)**	**Root DW ($mg.plant^{-1}$)**
None	Adequate	82 ± 3	38 ± 7
	Moderate Tox.	78 ± 9	30 ± 4
	Severe Tox.	82 ± 7	20 ± 3
Low	Adequate	77 ± 2	35 ± 6
	Moderate Tox.	81 ± 10	32 ± 10
	Severe Tox.	70 ± 12	21 ± 7
Medium	Adequate	78 ± 6	33 ± 5
	Moderate Tox.	71 ± 5	30 ± 6
	Severe Tox.	72 ± 8	25 ± 6
High	Adequate	64 ± 4	29 ± 5
	Moderate Tox.	71 ± 7	31 ± 5
	Severe Tox.	68 ± 9	25 ± 5

Shoot DW $HSD_{0.05}$ (S; BS; SxBS) = (8; *n.s.*; *n.s.*)
Root DW $HSD_{0.05}$ (S; BS; SxBS) = (*n.s.*; 5; *n.s.*)

Salinity, B toxicity and the interaction of these factors significantly affected the shoot B concentration and content (Table 3.1). Moderate and severe B toxicity treatments caused pronounced increases in shoot B concentration (Figure 3.3.A). However, increases of shoot B concentration under moderately and severely B-toxic conditions were limited by higher levels of salinity. Similar results were obtained for shoot B content (Figure 3.3.B). Boron content, which increased in the shoot with the application of moderate and severe B toxicity, decreased with increasing salinity, even more profoundly than B concentration.

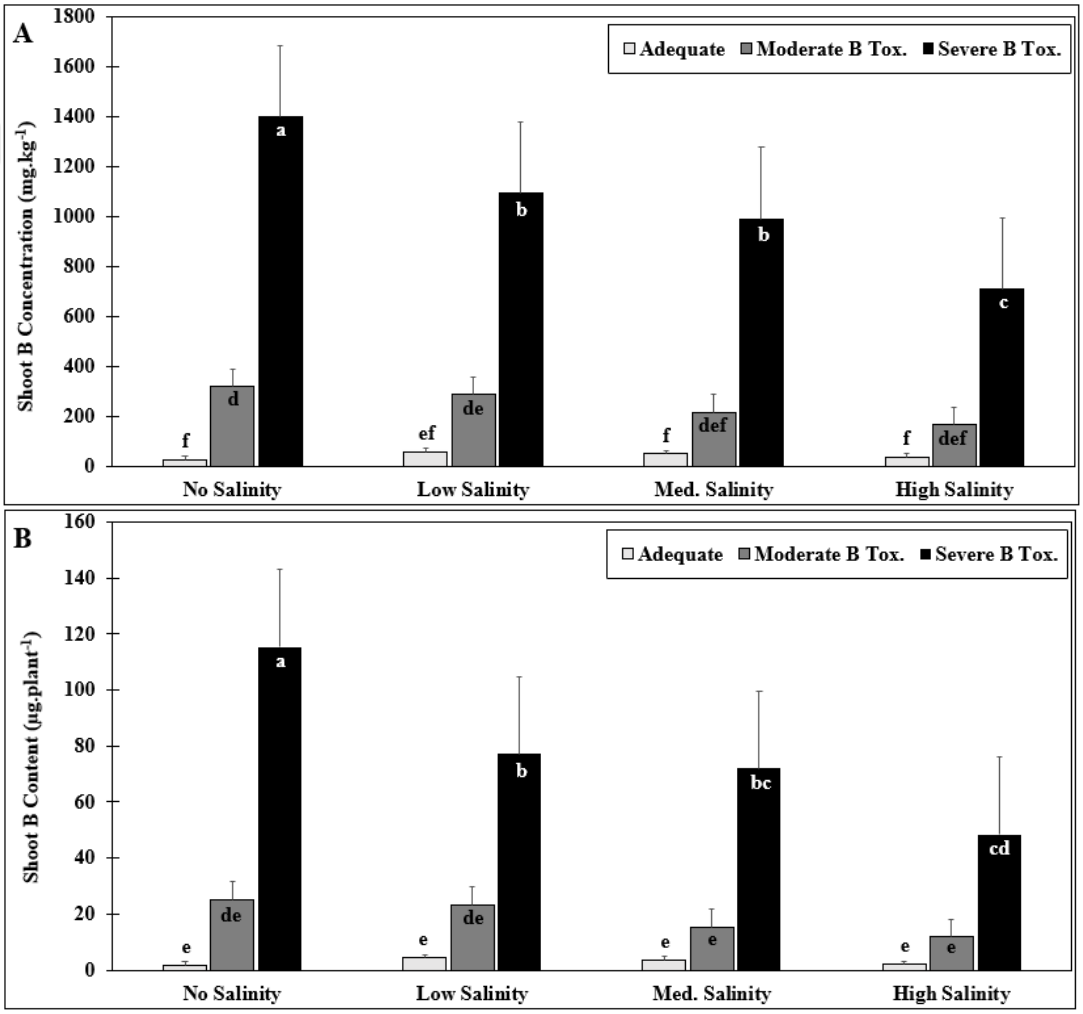

Figure 3.3: Effects of various doses of salinity (none: 0 mM NaCl; low: 40 mM NaCl; medium: 80 mM NaCl; high: 120 mM NaCl) and B supply (adequate: 2 µM; moderate toxicity: 0.5 mM; severe toxicity: 2.5 mM) on shoot B concentrations and contents of 4-weeks-old bread wheat (*Triticum aestivum* cv. Nusrat) grown hydroponically under growth chamber conditions. Different letters indicate significant differences between means according to two-way ANOVA and Tukey's HSD test ($p \leq 0.05$)

Moderate and severe B toxicity application to hydroponically grown wheat significantly elevated root B concentration and content (Table 3.1; Table 3.3). It was observed that in contrast to its effects on shoot B, salinity did not decrease the root B concentration and content in plants grown under B-toxic conditions but tended to increase these values under severe B toxicity.

Table 3.3: Effects of various levels of salinity (none: 0 mM NaCl; low: 40 mM NaCl; medium: 80 mM NaCl; high: 120 mM NaCl) and B supply (adequate: 2 µM; moderate toxicity: 0.5 mM; severe toxicity: 2.5 mM) on root B concentrations and contents of 4-weeks-old bread wheat (*Triticum aestivum* cv. Nusrat) grown hydroponically under growth chamber conditions. Different letters indicate significant differences between means according to two-way ANOVA and Tukey's HSD test ($p \leq 0.05$).

Salinity	**B Supply**	**Root B Conc. ($mg.kg^{-1}$)**	**Root B Content ($\mu g.plant^{-1}$)**
None	Adequate	17 d	0.62 c
	Moderate Tox.	26 cd	0.79 bc
	Severe Tox	39 bc	0.81 abc
Low	Adequate	13 d	0.43 c
	Moderate Tox.	20 cd	0.69 c
	Severe Tox	48 ab	0.99 abc
Medium	Adequate	7 d	0.22 c
	Moderate Tox.	19 cd	0.58 c
	Severe Tox	65 a	1.70 a
High	Adequate	8 d	0.22 c
	Moderate Tox.	16 d	0.49 c
	Severe Tox	66 a	1.68 ab

Although the salinity and B toxicity interaction had no significant effect on the shoot Na concentration, salinity significantly increased it whereas severe B toxicity had a limiting effect (Table 3.1; Table 3.4). The root Na concentration also increased markedly in response to salinity treatments, but it did not respond to B toxicity or B x salinity interaction. While the low and medium salinity treatments resulted in striking increases in shoot and root Na contents, the high salinity treatment did not further increase them.

Table 3.4: Shoot and root Na concentrations and contents of 4-weeks-old bread wheat (*Triticum aestivum* cv. Nusrat) grown hydroponically at different levels of B supply (adequate: 2 µM; moderate toxicity: 0.5 mM; severe toxicity: 2.5 mM), and salinity stress (none: 0 mM NaCl; low: 40 mM NaCl; medium: 80 mM NaCl; high: 120 mM NaCl). Data points represents the means and standard deviations of four replicates.

Salinity	**B Supply**	**Shoot Na Conc. (%)**	**Shoot Na Content (µg.plant^{-1})**	**Root Na Conc. (%)**	**Root Na Content (µg.plant^{-1})**
None	Adequate B	0.01 ± 0.00	10 ± 3	0.05 ± 0.03	17 ± 11
	Mod. Tox.	0.02 ± 0.01	14 ± 9	0.05 ± 0.03	15 ± 8
	Severe Tox.	0.01 ± 0.00	7 ± 2	0.04 ± 0.01	8 ± 2
Low	Adequate B	0.61 ± 0.10	468 ± 68	0.50 ± 0.11	176 ± 56
	Mod. Tox.	0.69 ± 0.14	558 ± 145	0.50 ± 0.19	176 ± 124
	Severe Tox.	0.54 ± 0.18	375 ± 106	0.50 ± 0.24	110 ± 75
Medium	Adequate B	1.05 ± 0.13	820 ± 142	1.00 ± 0.18	329 ± 64
	Mod. Tox.	0.97 ± 0.07	690 ± 47	1.01 ± 0.27	300 ± 98
	Severe Tox.	0.88 ± 0.10	630 ± 37	1.04 ± 0.31	272 ± 123
High	Adequate B	1.21 ± 0.04	776 ± 69	1.03 ± 0.21	307 ± 100
	Mod. Tox.	1.23 ± 0.11	876 ± 52	0.94 ± 0.15	292 ± 94
	Severe Tox.	1.13 ± 0.15	754 ± 27	1.33 ± 0.16	332 ± 36

Shoot Na Concentration $HSD_{0.05}$ (S; BS; SxBS)= (0.11; 0.07; *n.s.*)
Shoot Na Content $HSD_{0.05}$ (S; BS; SxBS)= (83; 65; 186)
Root Na Concentration $HSD_{0.05}$ (S; BS; SxBS)= (0.15; *n.s.*; *n.s.*)
Root Na Content $HSD_{0.05}$ (S; BS; SxBS)= (86; *n.s.*; *n.s.*)

Significant but modest decreases in shoot K concentrations and contents were observed upon salinity treatments irrespective of B toxicity (Table 3.1; Table 3.5). In contrast, root K levels were unaffected by salinity. However, in the absence of salinity stress, both the root K concentration as well as content exhibited a distinct decline with increasing severity of B toxicity.

Table 3.5: Effects of various levels of salinity (none: 0 mM NaCl; low: 40 mM NaCl; medium: 80 mM NaCl; high: 120 mM NaCl) and B supply (adequate: 2 µM; moderate toxicity: 0.5 mM; severe toxicity: 2.5 mM) on shoot and root K concentrations and contents of 4-weeks-old bread wheat (*Triticum aestivum* cv. Nusrat) grown hydroponically under growth chamber conditions. Data points represents the means and standard deviations of four replicates.

Salinity	**B Supply**	**Shoot K Conc. (%)**	**Shoot K Content (mg.plant^{-1})**	**Root K Conc. (%)**	**Root K Content (mg.plant^{-1})**
None	Adequate B	4.3 ± 0.3	3.5 ± 0.3	3.1 ± 0.8	1.2 ± 0.4
	Mod. Tox.	4.1 ± 0.2	3.2 ± 0.5	2.7 ± 0.7	0.8 ± 0.3
	Severe Tox.	4.3 ± 0.1	3.6 ± 0.4	1.5 ± 0.1	0.3 ± 0.0
Low	Adequate B	4.0 ± 0.1	3.1 ± 0.0	2.7 ± 0.6	1.0 ± 0.3
	Mod. Tox.	4.1 ± 0.2	3.3 ± 0.3	2.4 ± 1.0	0.8 ± 0.6
	Severe Tox.	4.2 ± 0.3	2.9 ± 0.6	2.5 ± 1.4	0.6 ± 0.4
Medium	Adequate B	4.1 ± 0.2	3.2 ± 0.4	2.8 ± 0.2	0.9 ± 0.1
	Mod. Tox.	4.0 ± 0.2	2.8 ± 0.2	2.8 ± 0.8	0.8 ± 0.3
	Severe Tox.	4.2 ± 0.1	3.1 ± 0.4	2.9 ± 1.1	0.8 ± 0.4
High	Adequate B	3.8 ± 0.1	2.5 ± 0.2	1.9 ± 0.6	0.6 ± 0.2
	Mod. Tox.	3.9 ± 0.1	2.8 ± 0.3	1.7 ± 0.5	0.5 ± 0.3
	Severe Tox.	4.0 ± 0.0	2.7 ± 0.3	2.9 ± 0.4	0.7 ± 0.1

Shoot K Concentration $HSD_{0.05}$ (S; BS; SxBS)= (0.2; 0.2; *n.s.*)
Shoot K Content $HSD_{0.05}$ (S; BS; SxBS)= (0.4; *n.s.*; *n.s.*)
Root K Concentration $HSD_{0.05}$ (S; BS; SxBS)= (*n.s.*; *n.s.*; 1.9)
Root K Content $HSD_{0.05}$ (S; BS; SxBS)= (*n.s.*; 0.3; *n.s.*)

In the statistical analysis of shoot and root K/Na ratios, the "no salinity" treatment group was left out because in the absence of any NaCl application, the nutrient solutions were almost Na-free and the measured tissue K concentrations were at least two orders of magnitude higher than the extremely low and variable tissue Na concentrations (data not shown). Increasing levels of salinity were associated with significantly lower K/Na ratios in both shoots and roots, irrespective of the B treatments (Table 3.1; Table 3.6).

Table 3.6: Shoot and root K/Na ratios of 4-weeks-old bread wheat (*Triticum aestivum* cv. Nusrat) grown hydroponically at different levels of B supply (adequate: 2 μM; moderate toxicity: 0.5 mM; severe toxicity: 2.5 mM), and salinity stress (none: 0 mM NaCl; low: 40 mM NaCl; medium: 80 mM NaCl; high: 120 mM NaCl). Data points represents the means and standard deviations of four replicates.

Salinity	B Supply	Shoot K/Na Ratio	Root K/Na Ratio
None	Adequate B	371 ± 78	92 ± 62
	Mod. Tox.	415 ± 200	94 ± 67
	Severe Tox.	562 ± 153	42 ± 12
Low	Adequate B	7 ± 1	6 ± 0
	Mod. Tox.	6 ± 2	5 ± 0
	Severe Tox.	8 ± 2	5 ± 1
Medium	Adequate B	4 ± 0	3 ± 0
	Mod. Tox.	4 ± 0	3 ± 0
	Severe Tox.	5 ± 1	3 ± 0
High	Adequate B	3 ± 0	2 ± 0
	Mod. Tox.	3 ± 0	2 ± 0
	Severe Tox.	4 ± 0	2 ± 0

Shoot K/Na Ratio $HSD_{0.05}$ (S; BT; SxBT)= (80; *n.s.*; *n.s.*)
Root K/Na Ratio $HSD_{0.05}$ (S; BT; SxBT)= (30; *n.s.*; *n.s.*)

Increasing salinity and B toxicity practices were reduced significantly shoot Ca concentration (Table 3.1; Table 3.7). It was observed that the root Ca concentration decreased in the presence of salinity stress. Although there was no distinction between salinity doses, it was observed that the root Ca concentration decreased in the presence of salinity.

The highest shoot Mg concentration in wheat were detected in plants grown under non-saline and adequate B conditions (Table 3.7). High salinity and both two B toxicity treatments, individually decreased the shoot and root Mg concentrations. Under high saline conditions, the impact of B toxicity to reduce root Mg concentration was disappeared.

Table 3.7: Effects of various levels of salinity (none: 0 mM NaCl; low: 40 mM NaCl; medium: 80 mM NaCl; high: 120 mM NaCl) and B supply (adequate: 2 µM; moderate toxicity: 0.5 mM; severe toxicity: 2.5 mM) on shoot and root Ca and Mg concentrations of 4-weeks-old bread wheat (*Triticum aestivum* cv. Nusrat) grown hydroponically under growth chamber conditions. Data points represents the means and standard deviations of four replicates.

Salinity	**B Supply**	**Shoot Ca Conc. (%)**	**Root Ca Conc. (%)**	**Shoot Mg Conc. (%)**	**Root Mg Conc. ($mg.kg^{-1}$)**
None	Adequate B	0.63 ± 0.05	0.15 ± 0.01	0.29 ± 0.01	924 ± 210
	Mod. Tox.	0.58 ± 0.02	0.15 ± 0.02	0.25 ± 0.02	762 ± 165
	Severe Tox.	0.52 ± 0.02	0.16 ± 0.03	0.23 ± 0.01	498 ± 40
Low	Adequate B	0.46 ± 0.04	0.10 ± 0.01	0.27 ± 0.02	983 ± 199
	Mod. Tox.	0.41 ± 0.03	0.10 ± 0.01	0.23 ± 0.02	750 ± 206
	Severe Tox.	0.35 ± 0.02	0.11 ± 0.00	0.19 ± 0.02	609 ± 237
Medium	Adequate B	0.36 ± 0.04	0.09 ± 0.00	0.24 ± 0.03	890 ± 88
	Mod. Tox.	0.34 ± 0.03	0.09 ± 0.01	0.21 ± 0.01	777 ± 126
	Severe Tox.	0.30 ± 0.01	0.10 ± 0.02	0.18 ± 0.01	694 ± 161
High	Adequate B	0.32 ± 0.02	0.10 ± 0.01	0.21 ± 0.02	589 ± 94
	Mod. Tox.	0.29 ± 0.02	0.10 ± 0.01	0.19 ± 0.02	535 ± 57
	Severe Tox.	0.27 ± 0.01	0.08 ± 0.01	0.16 ± 0.01	572 ± 69

Shoot Ca Concentration $HSD_{0.05}$ (S; BT; SxBT)= (0.03; 0.02; *n.s.*)
Root Ca Concentration $HSD_{0.05}$ (S; BT; SxBT)= (0.01; *n.s.*; *n.s.*)
Shoot Mg Concentration $HSD_{0.05}$ (S; BT; SxBT)= (0.02; 0.02; *n.s.*)
Root Mg Concentration $HSD_{0.05}$ (S; BT; SxBT)= (167; 131; *n.s.*)

3.1.2. Second Hydroponic Experiment Results

The second experiment was conducted to study the effects of the interaction of salinity stress and B toxicity on growth, shoot and root accumulation of minerals, necrosis severity, and distribution of minerals on different leaf sections in bread wheat under solution culture conditions.

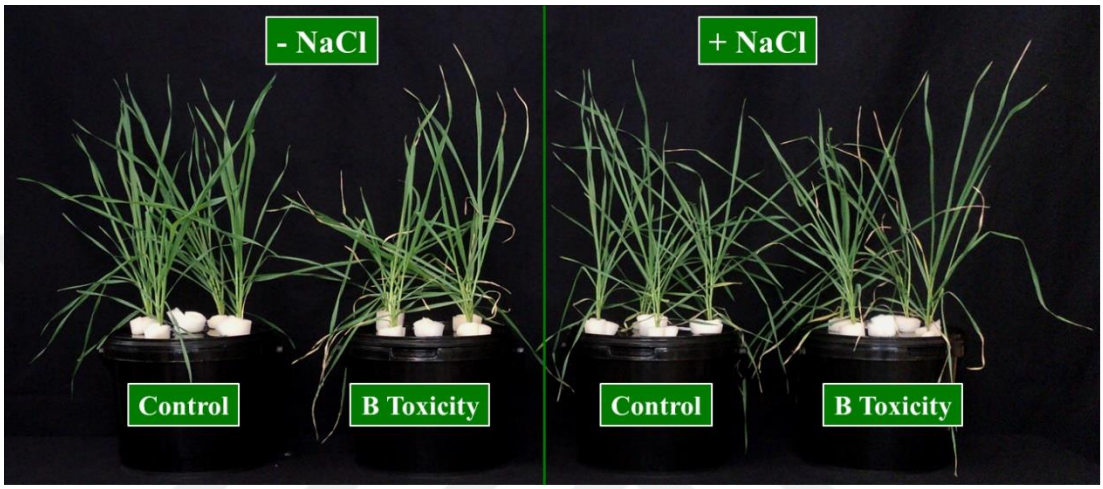

Figure 3.4: Shoot development of 4-weeks-old bread wheat (*Triticum aestivum* cv. Nusrat) grown hydroponically at two different levels of B supply (control: 2 μM; toxic: 1 mM), depending on absence or presence of salinity stress (-: 0 mM NaCl; +: 80 mM NaCl).

It was observed that the application of B toxicity to wheat plants caused the formation of necrosis and chlorosis on the tips and margins of the leaves (Figure 3.4). Salinity stress reduced the thickness of the control and B-toxic wheat leaves. Also, salinity stress was decreased necrosis intensity of leaves in B-toxic wheat plants (Figure 3.4; Figure 3.5).

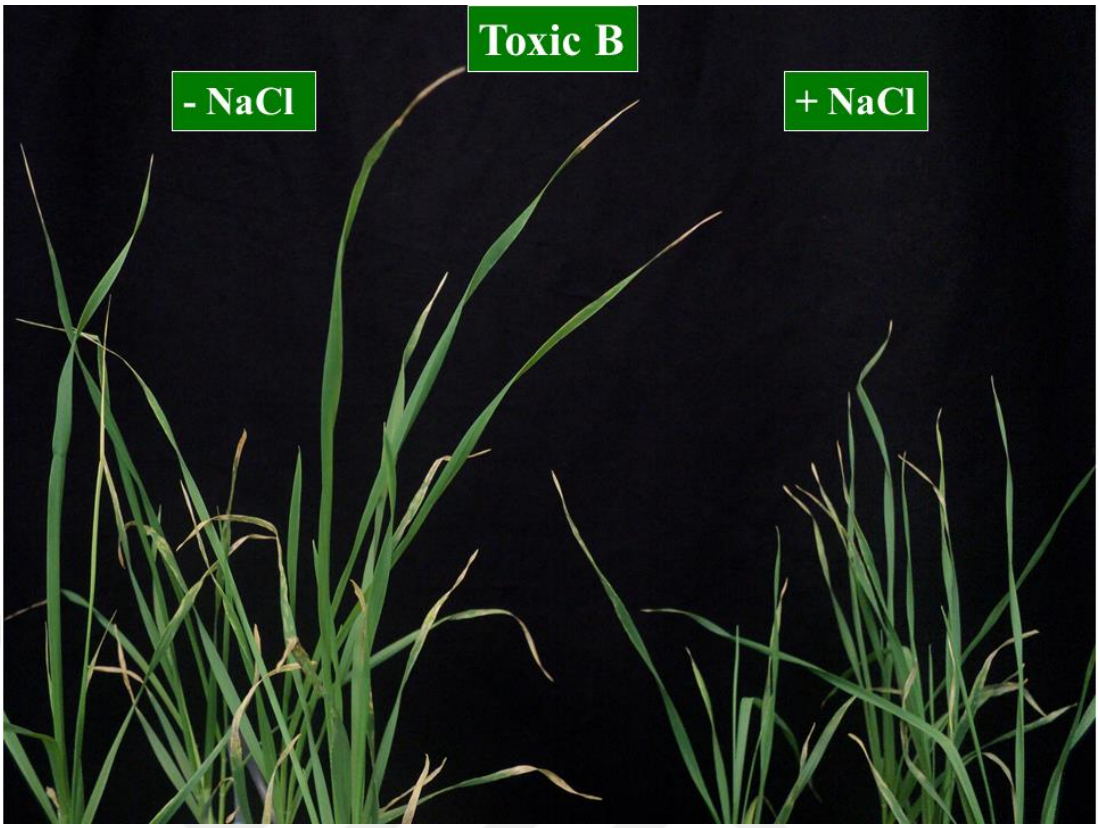

Figure 3.5: Effects of B toxicity (1 mM) on necrosis severity of 4-weeks-old bread wheat (*Triticum aestivum* cv. Nusrat) grown hydroponically at different levels of salinity (-: 0 mM NaCl; +: 80 mM NaCl) under growth chamber conditions.

Shoot DW was reduced significantly with the presence of salinity stress, but B toxicity had no major impact on shoot DW (Table 3.8; Table 3.9). In contrast, while salinity had no marked influence on root DW, B toxicity decrease significantly root DW.

Table 3.8: Shoot and root dry weights (DW) of 4-weeks-old bread wheat (*Triticum aestivum* cv. Nusrat) grown hydroponically at different levels of B supply (control: 2 µM; toxic: 1 mM), and salinity stress (-: 0 mM NaCl; +: 80 mM NaCl). Data points represents the means and standard deviations of four replicates.

Salinity	**Boron**	**Shoot DW (mg.plant^{-1})**	**Root DW (mg.plant^{-1})**
-NaCl	Control	132 ± 28	40 ± 12
	Toxic	141 ± 14	32 ± 6
+NaCl	Control	116 ± 16	42 ± 5
	Toxic	109 ± 18	34 ± 9

Shoot DW $LSD_{0.05}$ (S; BT; SxBT)= (21; *n.s.*; *n.s.*)
Root DW $LSD_{0.05}$ (S; BT; SxBT)= (*n.s.*; 6; *n.s.*)

Table 3.9: Analysis of variance of the main effects of salinity and B toxicity and their interaction on dry weights (DW), selected mineral concentrations, and K/Na ratios of shoots and roots of 4-weeks-old bread wheat (*Triticum aestivum* cv. Nusrat) grown hydroponically under growth chamber conditions.

Source of Variation	Shoot DW	Root DW	Shoot B Conc.	Root B Conc.
Salinity (S)	**	*n.s.*	**	**
B Toxicity (BT)	*n.s.*	***	***	***
S*BT	*n.s.*	*n.s.*	**	*n.s.*
Source of Variation	**Shoot Na Conc.**	**Root Na Conc.**	**Shoot K Conc.**	**Root K Conc.**
Salinity (S)	***	*	**	*
B Toxicity (BT)	*	*n.s.*	*n.s.*	*n.s.*
S*BT	*	*n.s.*	*n.s.*	*n.s.*
Source of Variation	**Shoot K/Na Ratio**	**Root K/Na Ratio**	**Shoot Ca Conc.**	**Root Ca Conc.**
Salinity (S)	***	**	***	**
B Toxicity (BT)	*n.s.*	*n.s.*	*n.s.*	*n.s.*
S*BT	*n.s.*	*n.s.*	*n.s.*	*n.s.*

n.s. Not significant; * 0.01 ≤ F Pr. < 0.05; ** 0.001 ≤ F Pr. < 0.01; *** F Pr. < 0.001

Salinity, B toxicity, and the interaction of these stress factors significantly affected the shoot B concentration (Table 3.9). Boron toxicity significantly increased the shoot B concentration (Table 3.10). But under toxic-B conditions, salinity stress reduced the shoot B concentration by 32%. Also, B toxicity significantly increased the root B concentration. However, root B concentration was reduced by salinity stress treatment to half.

Sodium concentration was improved significantly by salinity stress treatment (Table 3.9; Table 3.10). However, under saline conditions, B toxicity tended to reduce the shoot Na concentration. Also, root Na concentration increased in the presence of salinity stress.

The shoot and root K concentrations were reduced significantly in the presence of salinity in medium (Table 3.9; Table 3.10). But the main effect of B toxicity and combination with salinity stress were not affect significantly shoot and root K concentrations. While salinity significantly decreased shoot and root Ca concentrations, B toxicity treatment was found to have no significant effect (Table 3.9; Table 3.10).

Table 3.10: Effects of two different levels of salinity (-: 0 mM NaCl; +: 80 mM NaCl) and B supply (control: 2 μM; toxic: 1 mM) on the selected shoot and root mineral concentrations of 4-weeks-old bread wheat (*Triticum aestivum* cv. Nusrat) grown hydroponically under growth chamber conditions. Data points represents the means and standard deviations of four replicates.

Salinity	**B Toxicity**	**Shoot B Conc. (mg.kg-1)**	**Root B Conc. (mg.kg-1)**	**Shoot Na Conc. (%)**	**Root Na Conc. (%)**
-NaCl	Control	20 ± 5	15 ± 6	0.01 ± 0.00	0.02 ± 0.01
	Toxic	525 ± 44	43 ± 6	0.01 ± 0.00	0.02 ± 0.01
+NaCl	Control	32 ± 13	8 ± 2	1.45 ± 0.19	1.05 ± 0.12
	Toxic	397 ± 57	30 ± 5	1.15 ± 0.07	1.16 ± 0.15
Salinity	**B Toxicity**	**Shoot K Conc. (%)**	**Root K Conc. (%)**	**Shoot Ca Conc. (%)**	**Root Ca Conc. (%)**
-NaCl	Control	4.4 ± 0.1	2.6 ± 1.0	0.43 ± 0.03	0.17 ± 0.03
	Toxic	4.4 ± 0.1	2.9 ± 0.1	0.43 ± 0.01	0.17 ± 0.01
+NaCl	Control	4.1 ± 0.1	1.8 ± 0.3	0.29 ± 0.01	0.12 ± 0.00
	Toxic	4.2 ± 0.1	1.9 ± 0.4	0.25 ± 0.02	0.12 ± 0.01

Shoot B Concentration $LSD_{0.05}$ (S; BT; SxBT)= (40; 40; 78)
Root B Concentration $LSD_{0.05}$ (S; BT; SxBT)= (6; 6; *n.s.*)
Shoot Na Concentration $LSD_{0.05}$ (S; BT; SxBT)= (0.13; 0.13; 0.25)
Root Na Concentration $LSD_{0.05}$ (S; BT; SxBT)= (0.11; *n.s.*; *n.s.*)
Shoot K Concentration $LSD_{0.05}$ (S; BT; SxBT)= (0.1; *n.s.*; *n.s.*)
Root K Concentration $LSD_{0.05}$ (S; BT; SxBT)= (0.7; *n.s.*; *n.s.*)
Shoot Ca Concentration $LSD_{0.05}$ (S; BT; SxBT)= (0.02; *n.s.*; *n.s.*)
Root Ca Concentration $LSD_{0.05}$ (S; BT; SxBT)= (0.02; *n.s.*; *n.s.*)

In addition to the shoot B concentration (Table 3.10), B toxicity treatment also increased the shoot B content (Figure 3.6). However, salinity stress was decreased significantly shoot B content.

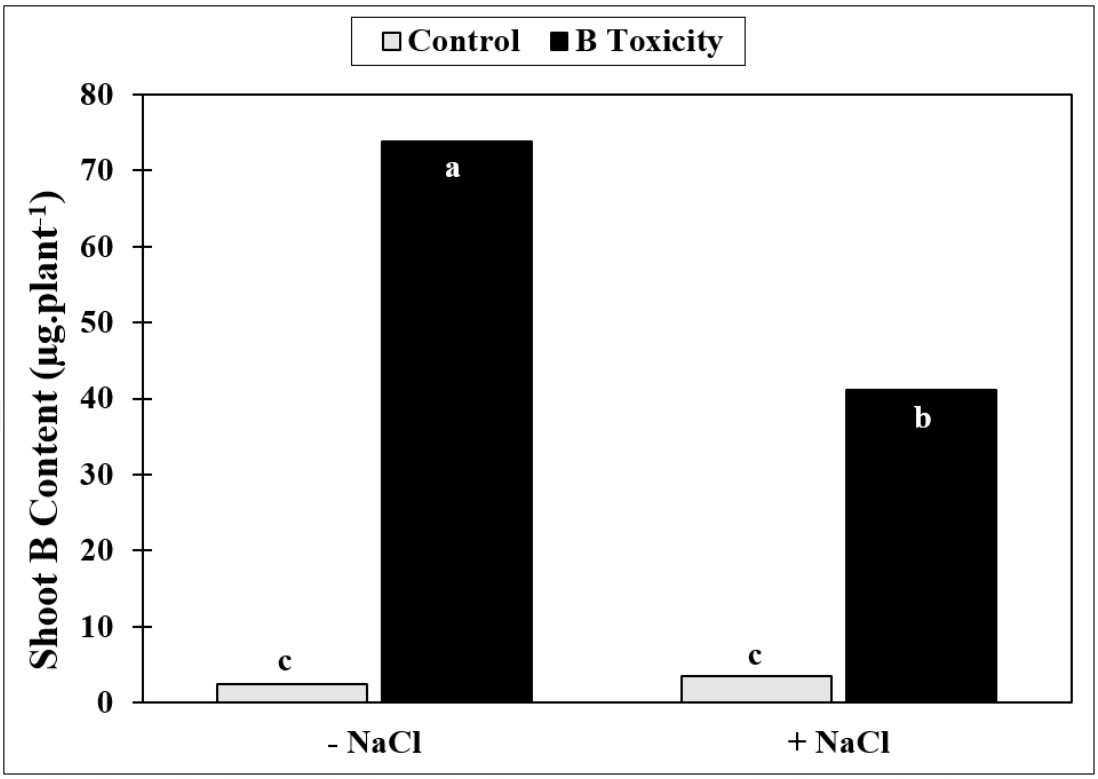

Figure 3.6: Shoot B content of 4-weeks-old bread wheat (*Triticum aestivum* cv. Nusrat) grown hydroponically at different levels of B supply (control: 2 µM; toxic: 1 mM), and salinity stress (-: 0 mM NaCl; +: 80 mM NaCl). Different letters indicate significant differences between means according to two-way ANOVA and Fisher's protected LSD test ($p \leq 0.05$).

Boron toxicity triggered necrosis formation in leaf tips and margins (Figure 3.5; Figure 3.7). However, it was observed that the necrosis severity shortened in the presence of salinity stress.

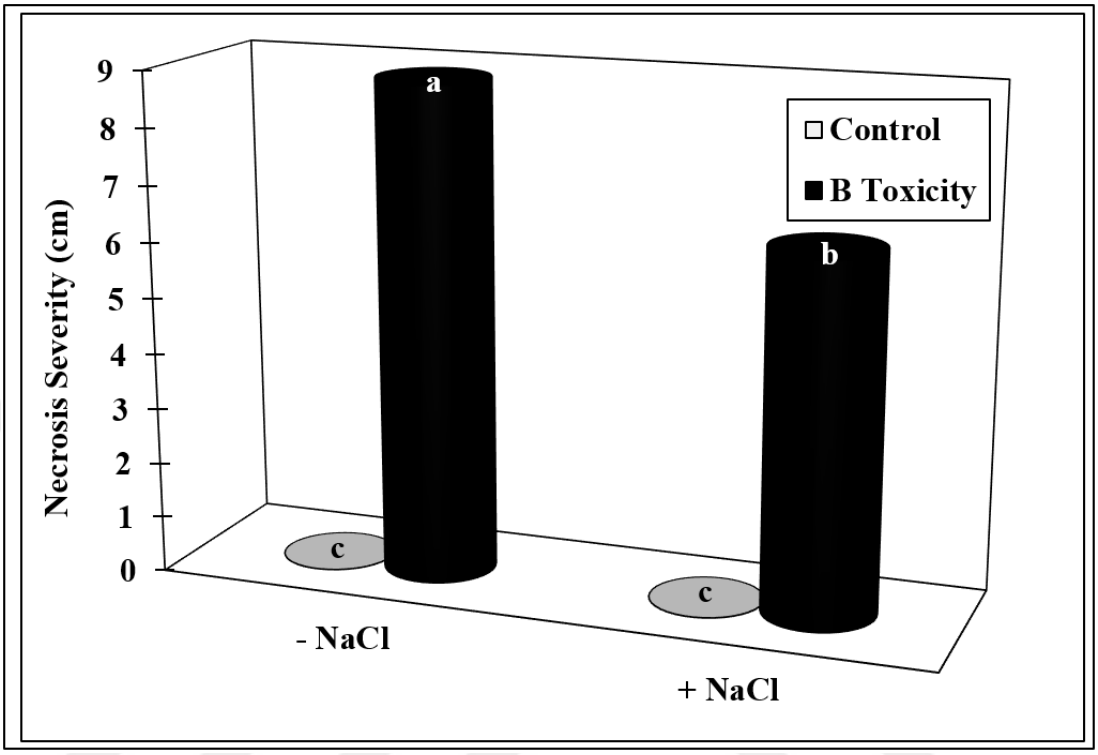

Figure 3.7: Effect of two different levels of salinity (-: 0 mM NaCl; +: 80 mM NaCl) and B supply (control: 2 μM; toxic: 1 mM) on necrosis severity of 4-weeks-old bread wheat (*Triticum aestivum* cv. Nusrat) grown hydroponically under growth chamber conditions. Different letters indicate significant differences between means according to two-way ANOVA and Fisher's protected LSD test ($p \leq 0.05$).

It was determined that shoot Na content was decreased significantly with increasing B toxicity under saline condition (Figure 3.8).

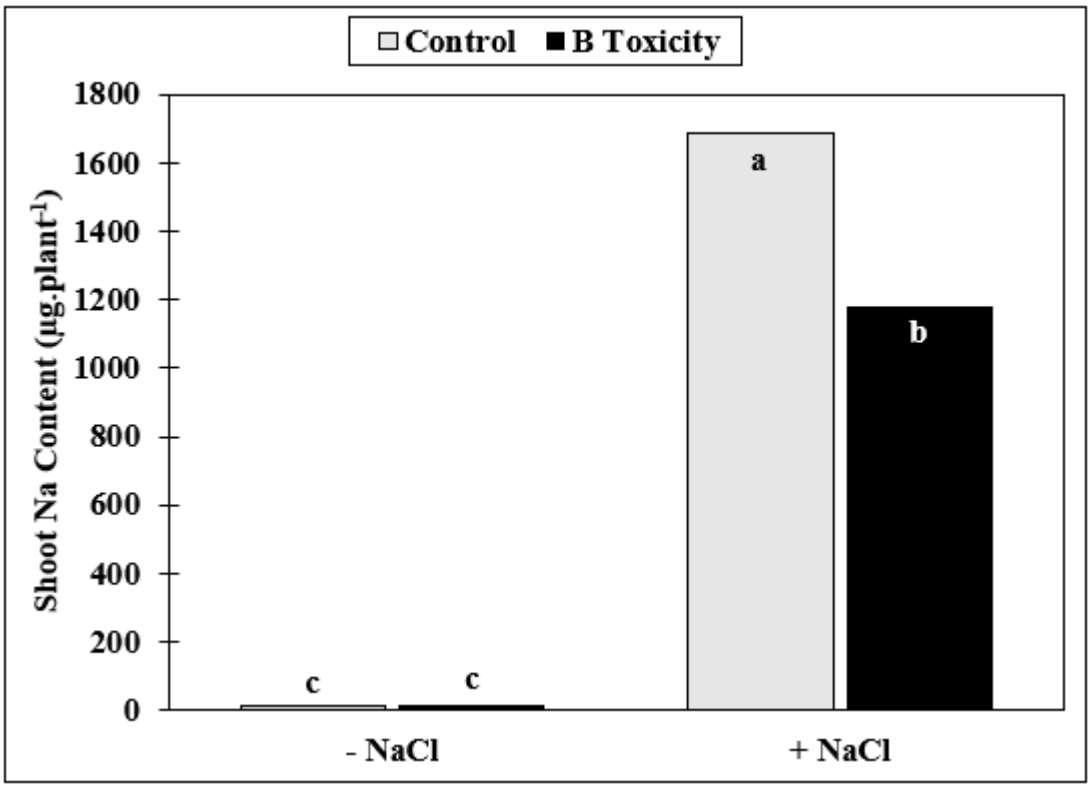

Figure 3.8: Shoot Na content of 4-weeks-old bread wheat (*Triticum aestivum* cv. Nusrat) grown hydroponically at different levels of B supply (control: 2 µM; toxic: 1 mM), and salinity stress (-: 0 mM NaCl; +: 80 mM NaCl). Different letters indicate significant differences between means according to two-way ANOVA and Fisher's protected LSD test ($p \leq 0.05$).

Wheat plants grown in non-saline media had high shoot and root K/Na ratios (Table 3.11). However, both shoot and root K/Na ratios were reduced significantly via the presence of salinity stress.

Table 3.11: Effect of two different levels of salinity (-: 0 mM NaCl; +: 80 mM NaCl) and B supply (control: 2 μM; toxic: 1 mM) on the shoot and root K/Na ratios of 4-weeks-old bread wheat (*Triticum aestivum* cv. Nusrat) grown hydroponically under growth chamber conditions. Data points represents the means and standard deviations of four replicates.

Salinity	**Boron**	**Shoot K/Na Ratio**	**Root K/Na Ratio**
-NaCl	Control	441 ± 190	166 ± 100
	Toxic	443 ± 42	151 ± 71
+NaCl	Control	2.9 ± 0.4	1.7 ± 0.1
	Toxic	3.6 ± 0.2	1.6 ± 0.1

Shoot K/Na Ratio $LSD_{0.05}$ (S; BT; SxBT)= (127; *n.s.*; *n.s.*)
Root K/Na Ratio $LSD_{0.05}$ (S; BT; SxBT)= (76; *n.s.*; *n.s.*)

The impact of B toxicity on the distribution of B, Na, and K among leaves of wheat plants under non-saline and saline conditions were shown (Table 3.12). The highest B concentration was determined in the necrotic parts of older leaves. However, salinity stress significantly decreased the B concentration in necrotic parts of older leaves. Boron concentration of the rest of the older leaves and young leaves were not affected by B toxicity. Sodium accumulation increased in all tissue types under saline conditions. It was determined that the K concentration in the necrotic parts of older leaves was lower than the young leaves and the rest of older leaves.

Table 3.12: Partitioning of B, Na, and K mineral nutrient concentrations among different tissue parts of 4-weeks-old bread wheat (*Triticum aestivum* cv. Nusrat) grown hydroponically at different levels of B supply (control: 2 µM; toxic: 1 mM), and salinity stress (-: 0 mM NaCl; +: 80 mM NaCl). Different letters indicate significant differences between means according to two-way ANOVA and Tukey's HSD test ($p \leq 0.05$).

Boron	**Salinity**	**Tissue Type**	**Mineral Concentrations**		
			B ($mg.kg^{-1}$)	**Na ($mg.kg^{-1}$)**	**K (%)**
High	Control	Young leaves	168 c	100 b	5.2 a
		Necrotic parts of older leaves	1506 a	133 b	2.8 c
		Rest of older leaves	106 c	60 b	4.8 ab
	High	Young leaves	108 c	2838 b	5.1 a
		Necrotic parts of older leaves	1103 b	16754 a	3.2 c
		Rest of older leaves	120 c	13530 a	4.1 b

3.1.3. Third Hydroponic Experiment Results

The third hydroponic experiment was conducted to evaluate the effects of the interaction of different levels of salinity stress with different levels of B toxicity on growth, leaf symptoms, shoot and root accumulation of minerals, transpiration rate, organic osmolyte accumulation, antioxidative enzyme activities, and oxidative damage in bread wheat and durum wheat under solution culture conditions.

Under both salinity conditions, B toxicity was caused chlorosis and necrosis in mature leaves of both wheat species beginning from leaf tips and margins and also inhibit the growth of plants (Figure 3.9). Moreover, the growth of plants was depressed by salinity stress. However, the necrosis severity of plants, which grow in high-saline conditions, was shorter than plants that are grown in non-saline media. When the general appearance of wheat species was compared, Saricanak seen to be well than Nusrat.

Boron toxicity treatment diminished root growth and expansion, especially in the cultivar of Saricanak (Figure 3.9). In addition, the root growth of two wheat species, especially in Nusrat, were depressed by salinity stress.

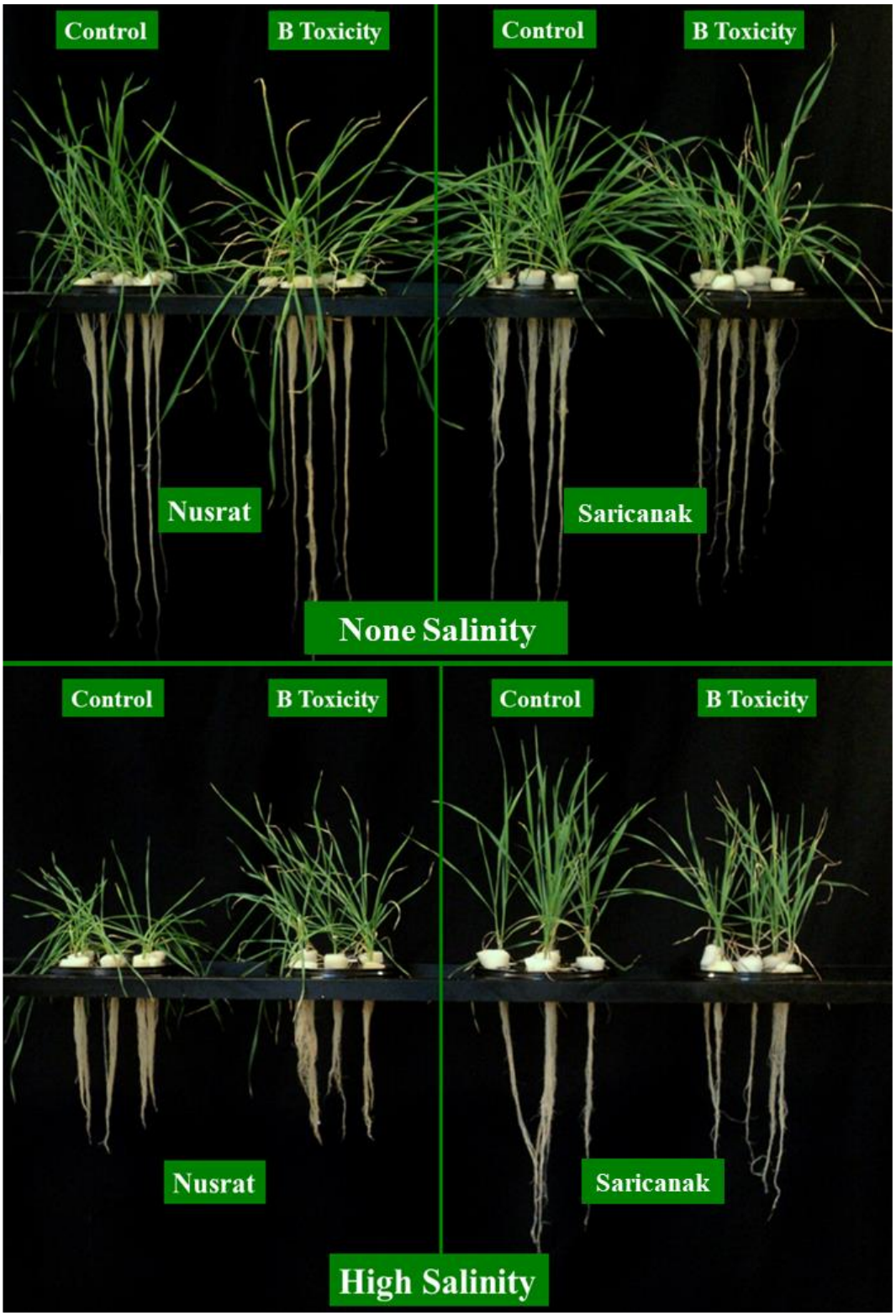

Figure 3.9: Shoot and root growth and general appearance of 4-weeks-old bread wheat (*Triticum aestivum* cv. Nusrat) and durum wheat (*Triticum durum* cv. Saricanak) grown hydroponically at two different levels of salinity stress (control: 0 mM NaCl; high: 120 mM NaCl), depending on absence or presence of B supply (control: 2 µM H_3BO_3; toxic: 1 mM H_3BO_3).

When leaves observed closely, the necrotic lesions were caused by B toxicity could be noticed clearly in two different wheat species under all three salinity conditions (Figure 3.10; Figure 3.11). But, increasing intensity of salinity decreased the necrosis severity of leaf tips and margins of both Nusrat and Saricanak cultivars.

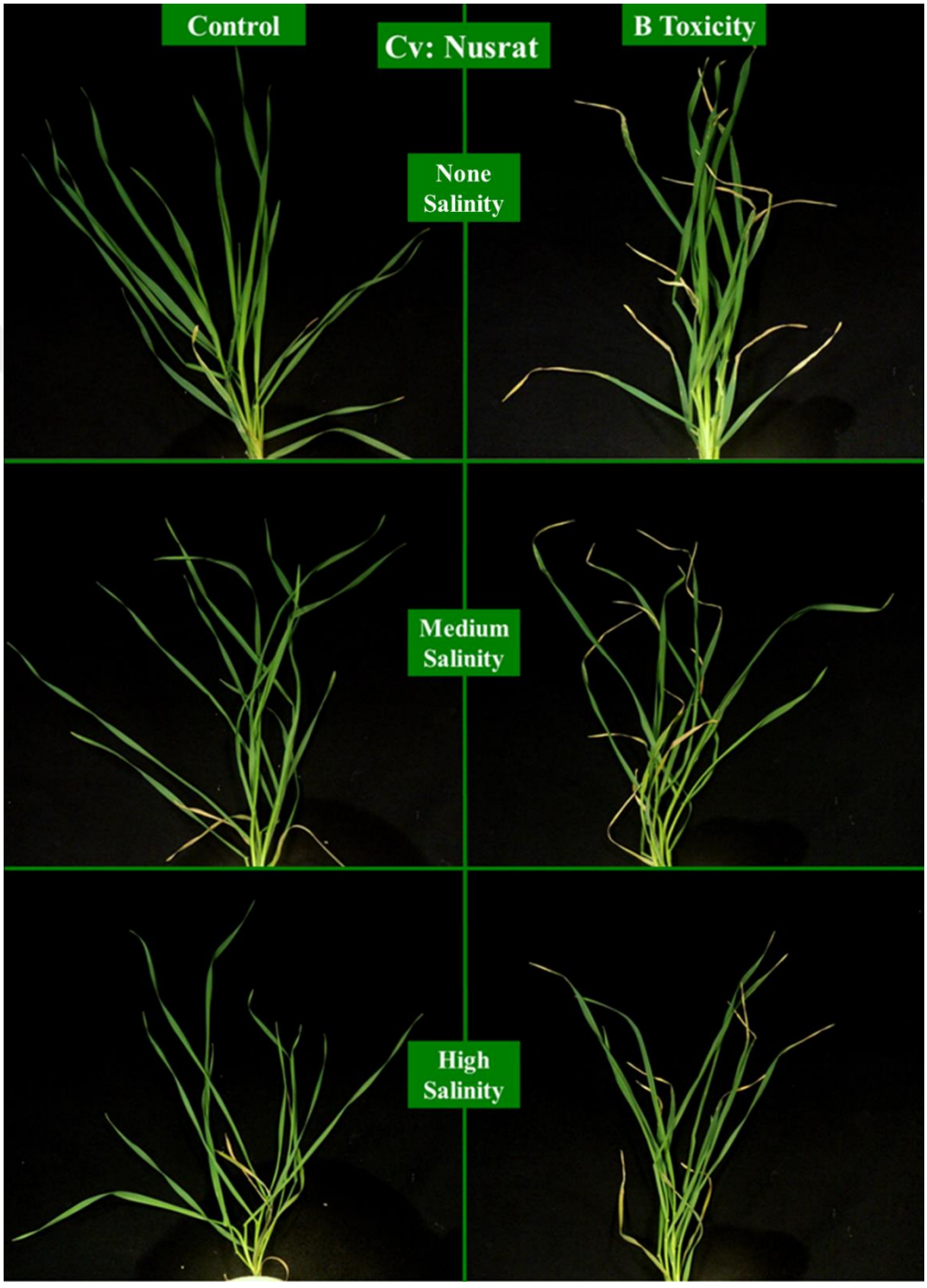

Figure 3.10: Close shot of leaves of 4-weeks-old bread wheat (*Triticum aestivum* cv. Nusrat) grown hydroponically at three different levels of salinity stress (control: 0 Mm NaCl; medium: 60 mM NaCl; high: 120 mM NaCl), depending on absence or presence of B supply (control: 2 µM H_3BO_3; toxic: 1 mM H_3BO_3).

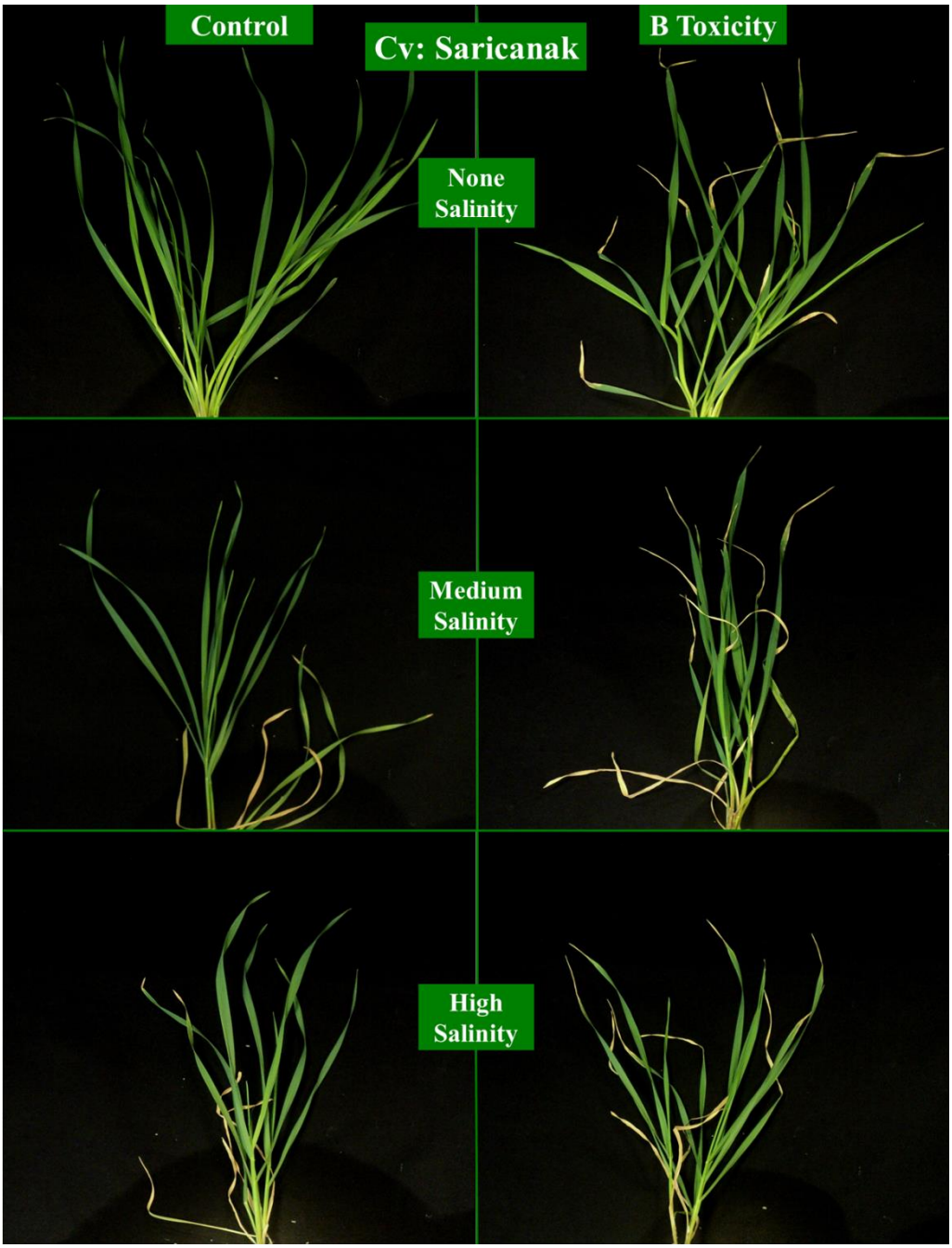

Figure 3.11: Close shot of leaves of 4-weeks-old durum wheat (*Triticum durum* cv. Saricanak) grown hydroponically at three different levels of salinity stress (control: 0 Mm NaCl; medium: 60 mM NaCl; high: 120 mM NaCl), depending on absence or presence of B supply (control: 2 μM H_3BO_3; toxic: 1 mM H_3BO_3).

Salt stress significantly reduced the shoot DW in both bread and durum wheat species (Table 3.13; Table 3.14). In addition, it was determined that durum wheat species had higher shoot DW than bread wheat under all stress conditions. On the other hand, B toxicity significantly reduced root DW in both Nusrat and Saricanak cultivars. In all situations, Nusrat was found to have a significantly higher root DW than Saricanak.

Table 3.13: Shoot and root dry weights (DW) of 4-weeks-old bread wheat (*Triticum aestivum* cv. Nusrat) and durum wheat (*Triticum durum* cv. Saricanak) grown hydroponically at different levels of salinity stress (control: 0 mM NaCl; medium: 60 mM NaCl; high 120 mM NaCl) and B supply (control: 2 µM; toxic: 1 mM). Data points represents the means and standard deviations of four replicates.

Cultivar	**Salinity**	**Boron**	**Shoot DW (mg.plant^{-1})**	**Root DW (mg.plant^{-1})**
Nusrat	None	Control	155 ± 20	51 ± 8
		Toxic	151 ± 27	39 ± 9
	Medium	Control	113 ± 25	49 ± 14
		Toxic	116 ± 17	41 ± 9
	High	Control	97 ± 23	50 ± 15
		Toxic	89 ± 8	39 ± 6
Saricanak	None	Control	218 ± 22	40 ± 3
		Toxic	183 ± 42	31 ± 9
	Medium	Control	157 ± 18	43 ± 8
		Toxic	147 ± 22	34 ± 6
	High	Control	120 ± 27	34 ± 11
		Toxic	116 ± 23	28 ± 8

Shoot DW $HSD_{0.05}$ (S; BT; CV; SxBT; BTxCV; SxCV; SxBTxCV)= (17; *n.s.*; 14; *n.s.*; *n.s.*; *n.s.*; *n.s.*)
Root DW $HSD_{0.05}$ (S; BT; CV; SxBT; BTxCV; SxCV; SxBTxCV)= (*n.s.*; 5; 5; *n.s.*; *n.s.*; *n.s.*; *n.s.*)

Table 3.14: Analysis of variance of the main effects of salinity and B toxicity and their interaction on dry weights (DW), selected mineral accumulations, and K/Na ratios of shoots and roots of 4-weeks-old bread wheat (*Triticum aestivum* cv. Nusrat) and durum wheat (*Triticum durum* cv. Saricanak) grown hydroponically under growth chamber conditions.

Source of Variation	Shoot DW	Root DW	Shoot K/Na Ratio	Root K/Na Ratio
Salinity (S)	***	*n.s.*	***	***
B Toxicity (BT)	*n.s.*	**	*n.s.*	*n.s.*
Cultivar (CV)	**	**	**	*n.s.*
S*BT	*n.s.*	*n.s.*	*n.s.*	*n.s.*
BT*CV	*n.s.*	*n.s.*	*n.s.*	*n.s.*
S*CV	*n.s.*	*n.s.*	**	*n.s.*
S*BT*CV	*n.s.*	*n.s.*	*n.s.*	*n.s.*
Source of Variation	**Shoot B Conc.**	**Shoot B Content**	**Root B Conc.**	**Root B Content**
Salinity (S)	***	***	***	***
B Toxicity (BT)	***	***	***	***
Cultivar (CV)	*	*n.s.*	*n.s.*	*
S*BT	***	***	***	*n.s.*
BT*CV	*	*n.s.*	**	*n.s.*
S*CV	*n.s.*	*n.s.*	**	*n.s.*
S*BT*CV	*n.s.*	*n.s.*	*n.s.*	*n.s.*
Source of Variation	**Shoot Na Conc.**	**Shoot Na Content**	**Root Na Conc.**	**Root Na Content**
Salinity (S)	***	***	***	***
B Toxicity (BT)	*n.s.*	*	*n.s.*	*
Cultivar (CV)	***	***	***	*n.s.*
S*BT	*n.s.*	*n.s.*	*n.s.*	*n.s.*
BT*CV	*n.s.*	*n.s.*	*n.s.*	*n.s.*
S*CV	***	***	***	*
S*BT*CV	*n.s.*	*n.s.*	*n.s.*	*n.s.*
Source of Variation	**Shoot K Conc.**	**Shoot Ca Conc.**	**Shoot P Conc.**	**Shoot Mg Conc.**
Salinity (S)	***	***	***	***
B Toxicity (BT)	*n.s.*	*n.s.*	*	***
Cultivar (CV)	***	*	**	***
S*BT	*n.s.*	*n.s.*	*n.s.*	*n.s.*
BT*CV	*n.s.*	*n.s.*	*n.s.*	*n.s.*
S*CV	***	*n.s.*	**	**
S*BT*CV	*n.s.*	*n.s.*	*n.s.*	*n.s.*
Source of Variation	**Shoot Fe Conc.**	**Shoot Zn Conc.**	**Shoot Mn Conc.**	**Shoot Cu Conc.**
Salinity (S)	***	***	***	***
B Toxicity (BT)	**	***	***	**
Cultivar (CV)	***	***	***	***
S*BT	*n.s.*	**	*n.s.*	*n.s.*
BT*CV	*n.s.*	**	*n.s.*	**
S*CV	**	***	*	**
S*BT*CV	*n.s.*	*n.s.*	*n.s.*	*n.s.*

n.s. Not significant; * $0.01 \leq F\ Pr. < 0.05$; ** $0.001 \leq F\ Pr. < 0.01$; *** $F\ Pr. < 0.001$

It was observed that the main effects of salinity, B toxicity, and the interaction of these two stress factors significantly affected shoot B concentration, shoot B content, and root B concentration (Table 3.14). In both species, the highest shoot B concentration, shoot B content, and root B concentration were determined in B-toxic non-saline wheat plants (Table 3.15). However, a significant decrease was detected in shoot B concentration, shoot B content, and root B concentration in plants treated with medium and high salinity stress under toxic-B conditions. Moreover, the root B content was increased in the presence of toxic-B, but salinity application tended to reduce it.

Table 3.15: Effects of different levels of salinity (control: 0 mM NaCl; medium: 60 mM NaCl; high 120 mM NaCl) and B supply (control: 2 μM; toxic: 1 mM) on the shoot and root B concentrations and contents of 4-weeks-old bread wheat (*Triticum aestivum* cv. Nusrat) and durum wheat (*Triticum durum* cv. Saricanak) grown hydroponically under growth chamber conditions. Data points represents the means and standard deviations of four replicates.

Cultivar	**Salinity**	**Boron**	**Shoot B Conc. ($mg.kg^{-1}$)**	**Shoot B Content ($\mu g.plant^{-1}$)**	**Root B Conc. ($mg.kg^{-1}$)**	**Root B Content ($\mu g.plant^{-1}$)**
Nusrat	None	Control	8 ± 1	1 ± 0	23 ± 11	1.1 ± 0.4
		Toxic	990 ± 83	151 ± 38	71 ± 8	2.8 ± 0.8
	Medium	Control	37 ± 16	5 ± 2	9 ± 2	0.4 ± 0.0
		Toxic	940 ± 57	110 ± 21	49 ± 7	2.0 ± 0.6
	High	Control	35 ± 18	3 ± 1	5 ± 1	0.3 ± 0.1
		Toxic	610 ± 33	55 ± 7	36 ± 4	1.4 ± 0.4
Saricanak	None	Control	28 ± 9	6 ± 1	4 ± 1	0.2 ± 0.0
		Toxic	924 ± 38	168 ± 36	70 ± 9	2.2 ± 0.9
	Medium	Control	36 ± 17	6 ± 2	5 ± 2	0.2 ± 0.1
		Toxic	850 ± 50	124 ± 30	61 ± 5	2.1 ± 0.5
	High	Control	30 ± 14	4 ± 1	5 ± 2	0.2 ± 0.0
		Toxic	610 ± 20	71 ± 15	42 ± 6	1.2 ± 0.5

Shoot B Conc. $HSD_{0.05}$ (S; BT; CV; SxBT; BTxCV; SxCV; SxBTxCV)= (32; 22; 22; 55; 40; n.*s.*; *n.s.*)
Shoot B Cont. $HSD_{0.05}$ (S; BT; CV; SxBT; BTxCV; SxCV; SxBTxCV)= (16; 11; *n.s.*; 29; *n.s.*; *n.s.*; *n.s.*)
Root B Conc. $HSD_{0.05}$ (S; BT; CV; SxBT; BTxCV; SxCV; SxBTxCV)= (5; 3; *n.s.*; 8; 6; 8; *n.s.*)
Root B Cont. $HSD_{0.05}$ (S; BT; CV; SxBT; BTxCV; SxCV; SxBTxCV)= (0.4; 0.2; 0.2; *n.s.*; *n.s.*; *n.s.*; *n.s.*)

Medium and high salinity stress significantly increased both shoot and root Na concentrations and contents (Table 3.14; Table 3.16). Although B toxicity significantly reduced shoot Na content in plants which exposed to salinity stress, it was determined that shoot Na concentration tended to decrease. Moreover, it was observed that B toxicity significantly decreased root Na content, but the same expression was not found on root Na concentration.

Cultivar difference significantly affected shoot Na concentration, shoot Na content, and root Na concentration (Table 3.14). Durum wheat cultivar accumulated more Na in shoot tissues than bread wheat variety (Table 3.16).

Table 3.16: Effects of different levels of salinity (control: 0 mM NaCl; medium: 60 mM NaCl; high 120 mM NaCl) and B supply (control: 2 µM; toxic: 1 mM) on the shoot and root Na concentrations and contents of 4-weeks-old bread wheat (*Triticum aestivum* cv. Nusrat) and durum wheat (*Triticum durum* cv. Saricanak) grown hydroponically under growth chamber conditions. Data points represents the means and standard deviations of four replicates.

Cultivar	**Salinity**	**Boron**	**Shoot Na Conc. (%)**	**Shoot Na Content (mg.plant^{-1})**	**Root Na Conc. (%)**	**Root Na Content (mg.plant^{-1})**
Nusrat	None	Control	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0
		Toxic	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0
	Medium	Control	2.1 ± 0.4	3.1 ± 1.2	2.0 ± 0.1	1.0 ± 0.3
		Toxic	2.0 ± 0.2	2.3 ± 0.4	2.1 ± 0.0	0.8 ± 0.2
	High	Control	2.6 ± 0.2	2.5 ± 0.8	2.6 ± 0.2	1.3 ± 0.5
		Toxic	2.7 ± 0.2	2.4 ± 0.4	2.6 ± 0.1	1.0 ± 0.2
Saricanak	None	Control	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0
		Toxic	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0
	Medium	Control	3.3 ± 0.4	5.1 ± 0.5	2.6 ± 0.0	1.1 ± 0.2
		Toxic	2.9 ± 0.4	4.3 ± 1.0	2.4 ± 0.1	0.8 ± 0.2
	High	Control	4.4 ± 0.5	5.3 ± 1.1	2.7 ± 0.1	0.9 ± 0.3
		Toxic	4.1 ± 0.3	4.7 ± 0.6	2.6 ± 0.1	0.7 ± 0.2

Shoot Na Conc. $HSD_{0.05}$ (S; BT; CV; SxBT; BTxCV; SxCV; SxBTxCV)= (0.2; *n.s.*; 0.2; *n.s.*; *n.s.*; 0.4; *n.s.*)
Shoot Na Cont. $HSD_{0.05}$ (S; BT; CV; SxBT; BTxCV; SxCV; SxBTxCV)= (0.5; 0.4; 0.4; *n.s.*; *n.s.*; 1; *n.s.*)
Root Na Conc. $HSD_{0.05}$ (S; BT; CV; SxBT; BTxCV; SxCV; SxBTxCV)= (0.08; *n.s.*; 0.05; *n.s.*; *n.s.*; 0.13; *n.s.*)
Root Na Cont. $HSD_{0.05}$ (S; BT; CV; SxBT; BTxCV; SxCV; SxBTxCV)= (0.2; 0.1; *n.s.*; *n.s.*; *n.s.*; 0.3; *n.s.*)

Medium and high salinity stress significantly reduced shoot K concentration (Table 3.14; Table 3.17). Shoot K concentration showed the difference between species under medium and high salinity conditions. According to this observation, shoot K concentration of bread wheat was determined higher than durum wheat.

Shoot Ca concentration was decreased significantly in the presence of salinity stress (Table 3.14; Table 3.17). Besides, it was determined that the shoot Ca concentration of the durum wheat was significantly higher than the bread wheat.

Medium and high salinity significantly enhanced the shoot P concentration in both bread and durum wheat species (Table 3.14; Table 3.17). A tendency to reduce shoot P concentration was detected in plants grown in B-toxic media. It was determined that the shoot P concentration of durum wheat was lower than the bread wheat plants except for non-saline application.

The main effects of salinity stress, B toxicity and interaction of them significantly reduced the shoot Mg concentration (Table 3.14; Table 3.17). Moreover, it was found that the shoot Mg concentration of Saricanak was lower than the Nusrat in each application of salinity stress and B toxicity, including the controls.

Table 3.17: Shoot K, Ca, P, and Mg macronutrients concentrations of 4-week-old bread wheat (*Triticum aestivum* cv. Nusrat) and durum wheat (*Triticum durum* cv. Saricanak) grown hydroponically at different levels of salinity (control: 0 mM NaCl; medium: 60 mM NaCl; high 120 mM NaCl) and B supply (control: 2 µM; toxic: 1 mM). Data points represents the means and standard deviations of four replicates.

Cultivar	**Salinity**	**Boron**	**Shoot K, Ca, P, and Mg Concentrations (%)**			
			K	**Ca**	**P**	**Mg**
Nusrat	None	Control	6.3 ± 0.1	0.54 ± 0.02	0.89 ± 0.02	0.27 ± 0.01
		Toxic	6.4 ± 0.2	0.54 ± 0.04	0.86 ± 0.03	0.25 ± 0.01
	Medium	Control	5.4 ± 0.3	0.32 ± 0.02	1.19 ± 0.07	0.27 ± 0.01
		Toxic	5.3 ± 0.2	0.30 ± 0.00	1.03 ± 0.06	0.22 ± 0.00
	High	Control	5.1 ± 0.3	0.25 ± 0.02	1.22 ± 0.11	0.20 ± 0.01
		Toxic	5.1 ± 0.3	0.24 ± 0.01	1.24 ± 0.08	0.18 ± 0.01
Saricanak	None	Control	6.1 ± 0.1	0.60 ± 0.02	0.89 ± 0.02	0.24 ± 0.02
		Toxic	6.4 ± 0.1	0.60 ± 0.02	0.93 ± 0.04	0.23 ± 0.02
	Medium	Control	4.3 ± 0.1	0.37 ± 0.02	1.00 ± 0.05	0.20 ± 0.01
		Toxic	4.3 ± 1.0	0.28 ± 0.19	0.92 ± 0.21	0.18 ± 0.03
	High	Control	3.9 ± 0.1	0.31 ± 0.01	1.14 ± 0.03	0.16 ± 0.00
		Toxic	3.9 ± 0.2	0.30 ± 0.01	1.06 ± 0.04	0.15 ± 0.01

Shoot K Conc. $HSD_{0.05}$ (S; BT; CV; SxBT; BTxCV; SxCV; SxBTxCV)= (0.3; *n.s.*; 0.2; *n.s.*; *n.s.*; 0.5; *n.s.*)
Shoot Ca Conc. $HSD_{0.05}$ (S; BT; CV; SxBT; BTxCV; SxCV; SxBTxCV)= (0.05; *n.s.*; 0.03; *n.s.*; *n.s.*; *n.s.*;)
Shoot P Conc. $HSD_{0.05}$ (S; BT; CV; SxBT; BTxCV; SxCV; SxBTxCV)= (0.07; 0.05; 0.05; *n.s.*; *n.s.*; 0.12; *n.s.*)
Shoot Mg Conc. $HSD_{0.05}$ (S; BT; CV; SxBT; BTxCV; SxCV; SxBTxCV)= (0.01; 0.01; 0.01; *n.s.*; *n.s.*; 0.02; *n.s.*)

Salinity stress significantly reduced shoot and root K/Na ratio in both bread and durum wheat species (Table 3.18). Also, according to the ANOVA results, the interaction between salinity and cultivar significantly affected the shoot K/Na ratio (Table 3.14). It was determined that Nusrat, which is a cultivar of bread wheat, had a higher shoot K/Na ratio than Saricanak which is a durum wheat variety in all stress treatment conditions, including controls (Table 3.18).

Table 3.18: Shoot and root K/Na ratio of 4-weeks-old bread wheat (*Triticum aestivum* cv. Nusrat) and durum wheat (*Triticum durum* cv. Saricanak) grown hydroponically at different levels of salinity (control: 0 mM NaCl; medium: 60 mM NaCl; high 120 mM NaCl) and B supply (control: 2 μM; toxic: 1 mM). Data points represents the means and standard deviations of four replicates.

Cultivar	**Salinity**	**Boron**	**Shoot K/Na Ratio**	**Root K/Na Ratio**
Nusrat	None	Control	3044 ± 2578	3208 ± 3143
		Toxic	2704 ± 1265	919 ± 440
	Medium	Control	2.6 ± 0.6	1.6 ± 0.1
		Toxic	2.7 ± 0.3	1.4 ± 0.0
	High	Control	2.0 ± 0.3	0.7 ± 0.0
		Toxic	1.9 ± 0.2	0.7 ± 0.0
Saricanak	None	Control	550 ± 99	618 ± 119
		Toxic	731 ± 135	1056 ± 730
	Medium	Control	1.3 ± 0.2	1.1 ± 0.1
		Toxic	1.5 ± 0.1	1.1 ± 0.1
	High	Control	0.9 ± 0.1	0.7 ± 0.0
		Toxic	1.0 ± 0.1	0.7 ± 0.0

Shoot K/Na ratio $HSD_{0.05}$ (S; BT; CV; SxBT; BTxCV; SxCV; SxBTxCV)= (597; *n.s.*; 484; *n.s.*; *n.s.*; 860; *n.s.*)
Root K/Na ratio $HSD_{0.05}$ (S; BT; CV; SxBT; BTxCV; SxCV; SxBTxCV)= (634; *n.s.*; *n.s.*; *n.s.*; *n.s.*; *n.s.*; *n.s.*;)

Irrespective of wheat species, the highest shoot Fe concentration was found in plants treated with medium salinity (Table 3.14; Table 3.19). In the control and high salinity stress applied plants, there was no difference observed in terms of shoot Fe concentration. On the other hand, B toxicity application to wheat plants lowered the shoot Fe concentration in general by 5%. Nusrat shoot Fe concentration was found to be significantly higher than Saricanak.

Shoot Zn concentration was significantly affected by salinity, B toxicity, cultivar, salinity*B interaction, B*cultivar interaction, and salinity*cultivar interaction (Table 3.14). The main effects of salinity and B toxicity significantly decreased shoot Zn concentration (Table 3.19). The shoot Zn concentration of Nusrat was found to be significantly higher than that of Saricanak.

The highest shoot Mn concentration was determined in the plants treated with medium salinity stress, while the lowest shoot Mn concentration was found in the high salinity applied wheat cultivars (Table 3.14 Table 3.19). High B toxicity reduced shoot Mn concentration by 25%. The shoot Mn concentration of Nusrat, which is a type of bread wheat, is higher than Saricanak in all salinity stress and B toxicity situations, including controls.

Whereas medium salinity stress did not affect shoot Cu concentration, high salinity treatment reduced shoot Cu concentration by 14% (Table 3.14; Table 3.19). Boron toxicity application tended to increase the shoot Cu concentration, especially in Saricanak. The shoot Cu concentration in Nusrat was 10% higher than that of Saricanak.

Table 3.19: Effects of different levels of salinity (control: 0 mM NaCl; medium: 60 mM NaCl; high 120 mM NaCl) and B supply (control: 2 µM; toxic: 1 mM) on shoot Fe, Zn, Mn, and Cu micronutrients concentrations of 4-weeks-old bread wheat (*Triticum aestivum* cv. Nusrat) and durum wheat (*Triticum durum* cv. Saricanak) grown hydroponically under growth chamber conditions. Data points represents the means and standard deviations of four replicates.

Cultivar	**Salinity**	**Boron**	**Shoot Fe, Zn, Mn, and Cu Concentrations (mg.kg^{-1})**			
			Fe	**Zn**	**Mn**	**Cu**
Nusrat	None	Control	90 ± 7	191 ± 16	322 ± 23	13.8 ± 0.9
		Toxic	81 ± 4	138 ± 11	278 ± 14	14.0 ± 0.6
	Medium	Control	84 ± 4	182 ± 18	357 ± 14	14.3 ± 0.5
		Toxic	84 ± 6	148 ± 6	306 ± 10	13.9 ± 0.7
	High	Control	80 ± 3	110 ± 6	248 ± 23	12.4 ± 0.4
		Toxic	78 ± 7	97 ± 2	236 ± 4	12.9 ± 0.2
Saricanak	None	Control	69 ± 2	100 ± 10	240 ± 14	12.7 ± 0.8
		Toxic	61 ± 5	80 ± 8	218 ± 16	14.0 ± 0.2
	Medium	Control	80 ± 2	93 ± 5	251 ± 19	12.0 ± 0.6
		Toxic	77 ± 3	81 ± 13	234 ± 6	13.3 ± 0.3
	High	Control	70 ± 2	79 ± 4	197 ± 7	10.6 ± 0.3
		Toxic	65 ± 4	70 ± 12	183 ± 20	11.1 ± 0.6

Shoot Fe Conc. $HSD_{0.05}$ (S; BT; CV; SxBT; BTxCV; SxCV; SxBTxCV)= (4; 3; 3; *n.s.*; *n.s.*; 7; *n.s.*)
Shoot Zn Conc. $HSD_{0.05}$ (S; BT; CV; SxBT; BTxCV; SxCV; SxBTxCV)= (9; 6; 6; 16; 11; 16; *n.s.*)
Shoot Mn Conc. $HSD_{0.05}$ (S; BT; CV; SxBT; BTxCV; SxCV; SxBTxCV)= (14; 9; 9; *n.s.*; *n.s.*; 24; *n.s.*)
Shoot Cu Conc. $HSD_{0.05}$ (S; BT; CV; SxBT; BTxCV; SxCV; SxBTxCV)= (0.5; 0.3; 0.3; *n.s.*; 0.6; 0.8; *n.s.*)

Boron toxicity caused necrosis formation in leaf tips and margins in both wheat species (Figure 3.10; Figure 3.11). However, in the presence of medium salinity, the severity of necrosis reduced, and this decrease was exacerbated when high salinity was applied (Figure 3.12). A difference was found between bread wheat and durum wheat in terms of reducing the effect of salinity on the necrosis severity. The necrosis severity of Nusrat was decreased more severely than Saricanak.

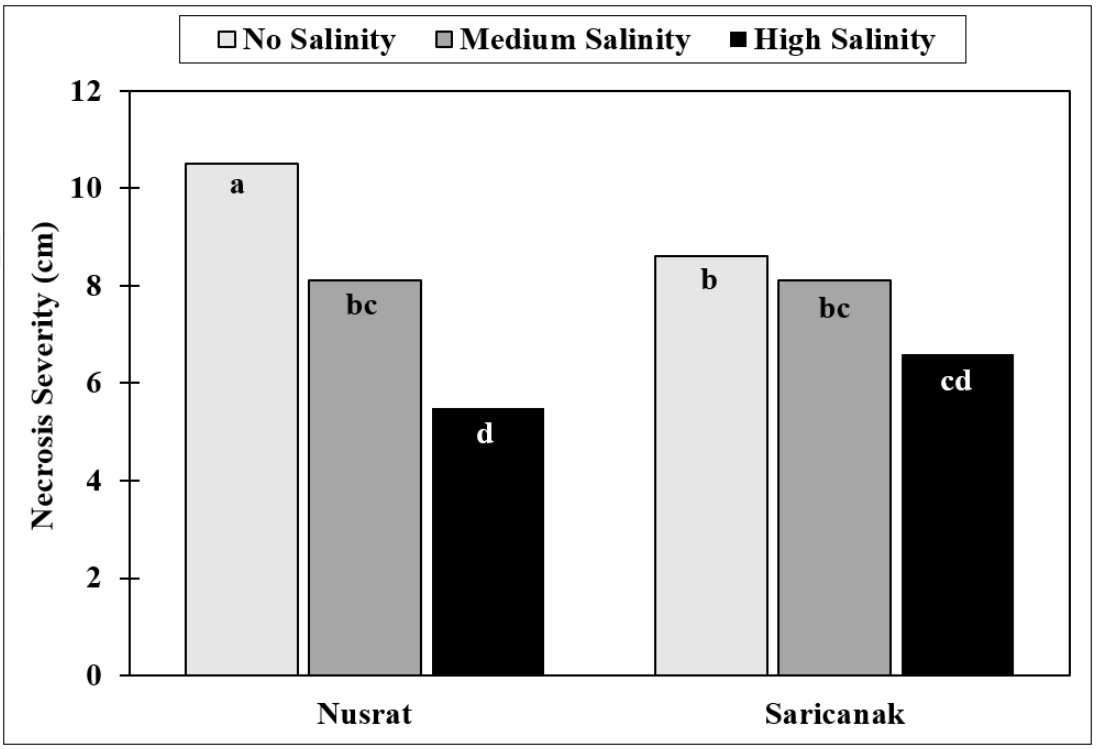

Figure 3.12: Effects of various levels of salinity (control: 0 mM NaCl; medium: 60 mM NaCl; high 120 mM NaCl) and B supply (control: 2 µM; toxic: 1 mM) on necrosis severity of 4-weeks-old bread wheat (*Triticum aestivum* cv. Nusrat) and durum wheat (*Triticum durum* cv. Saricanak) grown hydroponically under growth chamber conditions. Different letters indicate significant differences between means according to two-way ANOVA and Tukey's HSD test ($p \leq 0.05$).

Table 3.20: Analysis of variance of the main effects of salinity and B toxicity and their interaction on transpiration rate, relative electrolyte leakage, concentrations of malondialdehyde (MDA), glycine betaine (GB), and proline, total and specific enzyme activities of superoxide dismutase (SOD), glutathione reductase (GR), ascorbate peroxidase (AP), and catalase (CAT) of shoots and roots of 4-weeks-old bread wheat (*Triticum aestivum* cv. Nusrat) and (*Triticum durum* cv. Saricanak) grown hydroponically under growth chamber conditions.

Source of Variation	**Transpiration Rate***			
	t_0	**t_1**	**t_2**	**t_3**
Salinity (S)	*n.s.*	***	***	***
B Toxicity (BT)	*n.s.*	*n.s.*	*	*
Cultivar (CV)	*	*n.s.*	*n.s.*	**
S*BT	*n.s.*	*n.s.*	*n.s.*	*
BT*CV	*n.s.*	*n.s.*	*n.s.*	*n.s.*
S*CV	*n.s.*	*n.s.*	*n.s.*	*
S*BT*CV	*n.s.*	*n.s.*	*n.s.*	*n.s.*
Source of Variation	**REL**	**MDA Conc.**	**GB Conc.**	**Proline Conc.**
Salinity (S)	**	***	***	**
B Toxicity (BT)	***	*n.s.*	*n.s.*	*n.s.*
Cultivar (CV)	*n.s.*	**	*	*n.s.*
S*BT	*n.s.*	*n.s.*	*n.s.*	*n.s.*
BT*CV	*n.s.*	*n.s.*	*n.s.*	*n.s.*
S*CV	*n.s.*	*n.s.*	**	*n.s.*
S*BT*CV	*	*n.s.*	*n.s.*	*n.s.*
Source of Variation	**SOD Act.**	**GR Act.**	**AP Act.**	**CAT Act.**
Salinity (S)	***	*n.s.*	**	*
B Toxicity (BT)	*n.s.*	*n.s.*	*n.s.*	*n.s.*
Cultivar (CV)	*	*n.s.*	**	*n.s.*
S*BT	*n.s.*	*n.s.*	*n.s.*	*n.s.*
BT*CV	*n.s.*	*n.s.*	*n.s.*	*n.s.*
S*CV	*n.s.*	*	**	**
S*BT*CV	*n.s.*	*n.s.*	*n.s.*	*n.s.*
Source of Variation	**Sp. SOD Act.**	**Sp. GR Act.**	**Sp. AP Act.**	**Sp. CAT Act.**
Salinity (S)	*n.s.*	*n.s.*	*n.s.*	*n.s.*
B Toxicity (BT)	*	*n.s.*	*n.s.*	*
Cultivar (CV)	*n.s.*	*	*n.s.*	**
S*BT	*n.s.*	*n.s.*	*n.s.*	*n.s.*
BT*CV	*n.s.*	*n.s.*	*n.s.*	*n.s.*
S*CV	*n.s.*	*n.s.*	*	***
S*BT*CV	*n.s.*	*n.s.*	*n.s.*	*n.s.*

*24^{th} hours transpiration rate: t_0: before stress application (10 DAS), t_1: after first stress application (14 DAS), t_2: after second stress application (21 DAS) and t_3: after third stress application (28 DAS)

As mentioned in the material method section, the transpiration rate was measured in plants at the 6th and 24th hours. Since the 6th and 24th hours transpiration rate results were consistent between each other and there was no statistically significant difference between them, the 6^{th}-hour data were not shown. (Table 3.20; Table 3.21).

The difference of cultivar significantly affected the results of transpiration rate at time t0, i.e. before stress application (Table 3.20). According to this result, it was determined that transpiration at t0 time was higher in durum wheat (Table 3.21). Furthermore, salinity significantly reduced transpiration rate in both bread and durum varieties at time t1, i.e. after the first application of stress. The main effects of salinity and B toxicity significantly reduced transpiration rate at t2, i.e. the second stress application period.

Salinity, B toxicity, cultivar, salinity*B toxicity, and salinity*cultivar interaction significantly affected the transpiration rate at t_3, i.e. after the 3rd stress application (Table 3.20). The main effects of B toxicity and especially salinity stress significantly reduced transpiration rate at t_3. Transpiration at t_3 was found to be 30% higher in durum wheat than in bread wheat.

Table 3.21: Effects of different levels of salinity (control: 0 mM NaCl; medium: 60 mM NaCl; high 120 mM NaCl) and B supply (control: 2 μM; toxic: 1 mM) on the transpiration rate of 4-weeks-old bread wheat (*Triticum aestivum* cv. Nusrat) and durum wheat (*Triticum durum* cv. Saricanak) grown hydroponically under growth chamber conditions. Data points represents the means and standard deviations of four replicates.

Cultivar	**Salinity**	**Boron**	**Transpiration (ml.plant.24^{-1})**			
			t_0	**t_1**	**t_2**	**t_3**
Nusrat	None	Control	2.8 ± 0.4	3.8 ± 0.3	7.1 ± 1.0	10.9 ± 1.0
		Toxic	2.8 ± 0.5	3.8 ± 0.4	5.9 ± 0.8	8.9 ± 1.5
	Medium	Control	2.8 ± 0.3	3.2 ± 0.6	4.9 ± 0.5	6.5 ± 1.2
		Toxic	2.8 ± 0.8	3.3 ± 0.3	4.3 ± 0.9	6.1 ± 1.0
	High	Control	2.8 ± 0.4	2.5 ± 0.2	3.4 ± 0.8	4.7 ± 1.2
		Toxic	3.3 ± 0.7	2.8 ± 0.3	3.5 ± 0.8	4.6 ± 0.8
Saricanak	None	Control	3.2 ± 0.4	3.8 ± 0.3	8.0 ± 0.7	17.1 ± 5.0
		Toxic	2.9 ± 0.5	3.8 ± 0.3	6.5 ± 1.3	12.0 ± 2.0
	Medium	Control	3.2 ± 0.3	3.5 ± 0.3	5.1 ± 0.8	8.2 ± 1.5
		Toxic	3.5 ± 0.3	3.8 ± 0.6	4.8 ± 0.3	7.6 ± 0.1
	High	Control	3.2 ± 0.3	2.4 ± 0.5	3.4 ± 0.4	5.1 ± 1.3
		Toxic	3.4 ± 0.6	2.6 ± 0.3	3.2 ± 0.5	4.4 ± 1.3

t_0 $HSD_{0.05}$ (S; BT; CV; SxBT; BTxCV; SxCV; SxBTxCV)= (*n.s.*; *n.s.*; 0.3; *n.s.*; *n.s.*; *n.s.*;)
t_1 $HSD_{0.05}$ (S; BT; CV; SxBT; BTxCV; SxCV; SxBTxCV)= (0.3; *n.s.*; *n.s.*; *n.s.*; *n.s.*; *n.s.*;)
t_2 $HSD_{0.05}$ (S; BT; CV; SxBT; BTxCV; SxCV; SxBTxCV)= (0.5; 0.4; *n.s.*; *n.s.*; *n.s.*; *n.s.*; *n.s.*)
t_3 $HSD_{0.05}$ (S; BT; CV; SxBT; BTxCV; SxCV; SxBTxCV)= (1.3; 1.1; 1.1; 1.9; *n.s.*; 1.9; *n.s.*)
*24^{th} hours transpiration: t_0: before stress application (10 DAS), t_1: after first stress application (14 DAS), t_2: after second stress application (21 DAS) and t_3: after third stress application (28 DAS)

Normalized transpiration rate after the t_3 period was decreased in both bread and durum wheat varieties as a result of salinity stress (Figure 3.13). It was determined that the normalized transpiration rate result of the plants under high salinity stress was lower than the plants were exposed to medium salinity. In addition, it was found that the normalized transpiration rate of Nusrat was lower than that of Saricanak under all stress conditions, except plants that were grown in high-saline media. On the other hand, no significant effect of B toxicity was revealed.

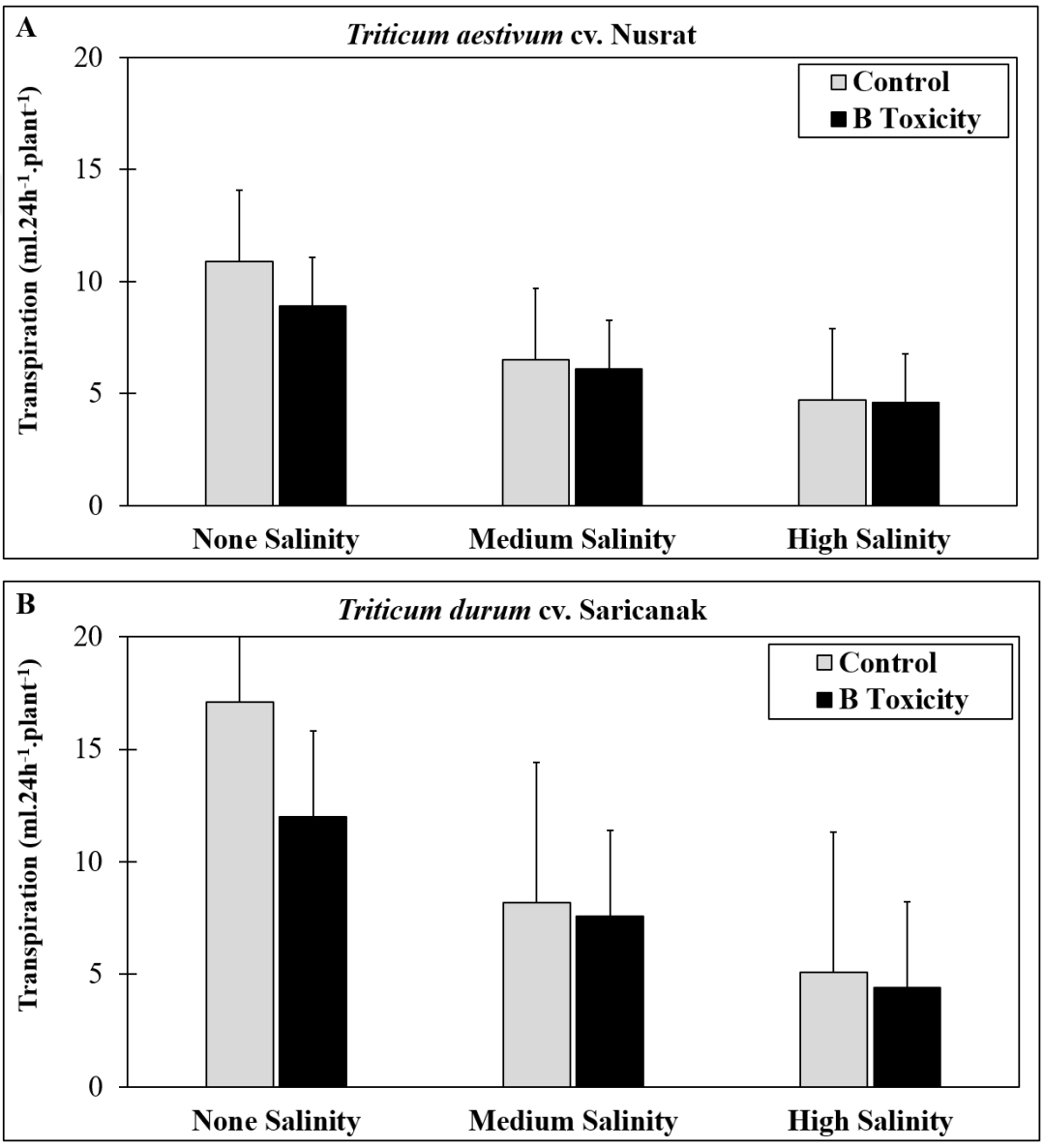

Figure 3.13: Normalized transpiration rate results after third stress application (t_3) of 4-weeks-old bread wheat (*Triticum aestivum* cv. Nusrat) and durum wheat (*Triticum durum* cv. Saricanak) grown hydroponically at different levels of salinity (control: 0 mM NaCl; medium: 60 mM NaCl; high 120 mM NaCl) and B supply (control: 2 µM; toxic: 1 mM). Bars represents standard deviations.

Medium and high salinity stress treatments increased relative electrolyte leakage (REL) by approximately 45% (Table 3.22). In addition, B toxicity significantly enhanced REL in both Nusrat and Saricanak cultivars under all salinity conditions, including control. According to the ANOVA results of REL, triple interaction between salinity, B toxicity, and cultivar was detected (Table 3.20).

Both medium and high salinity treatments significantly increased the malondialdehyde (MDA) concentration (Table 3.20; Table 3.22). Medium salinity increased MDA concentration by 275% compared to control, while high salinity increased by 350%. It was determined that the MDA concentration of Nusrat was significantly higher than Saricanak.

Table 3.22: Effects of different levels of salinity (control: 0 mM NaCl; medium: 60 mM NaCl; high 120 mM NaCl) and B supply (control: 2 μM; toxic: 1 mM) on relative electrolyte leakage (REL) of third-oldest leaves and malondialdehyde (MDA) concentrations of 4-weeks-old bread wheat (*Triticum aestivum* cv. Nusrat) and durum wheat (*Triticum durum* cv. Saricanak) grown hydroponically under growth chamber conditions. Data points represents the means and standard deviations of four replicates.

Cultivar	**Salinity**	**Boron**	**REL (nmol.ml^{-1})**	**MDA Conc. (nmol.ml^{-1})**
Nusrat	None	Control	5 ± 2	10 ± 3
		Toxic	24 ± 1	10 ± 1
	Medium	Control	11 ± 7	25 ± 11
		Toxic	25 ± 7	31 ± 10
	High	Control	11 ± 5	37 ± 8
		Toxic	21 ± 6	35 ± 12
Saricanak	None	Control	4 ± 2	6 ± 2
		Toxic	14 ± 3	13 ± 11
	Medium	Control	9 ± 3	18 ± 4
		Toxic	21 ± 2	17 ± 4
	High	Control	8 ± 1	20 ± 12
		Toxic	27 ± 7	25 ± 5

REL $HSD_{0.05}$ (S; BT; CV; SxBT; BTxCV; SxCV; SxBTxCV)= (4; 3.; *n.s.*; *n.s.*; *n.s.*; *n.s.*; 11)
MDA Conc. $HSD_{0.05}$ (S; BT; CV; SxBT; BTxCV; SxCV; SxBTxCV)= (6; *n.s.*; 4; *n.s.*; *n.s.*; *n.s.*; *n.s.*)

While main effect of B toxicity had no effect on glycine betaine (GB) concentration, a significant interaction was detected between salinity and cultivar (Table 3.20; Table 3.23). The lowest GB concentration was determined in the Saricanak under non-saline conditions. Medium and high salinity stress increased the GB concentration in Saricanak by 100% and there was not found statistically significant difference between them. When both wheat species were compared in salinity control, the concentration of GB in Nusrat was found 35% higher than in Saricanak. Medium salinity stress treatment did not increase Nusrat GB concentration, while high salinity enhanced GB concentration by 80%.

When considering all variables, just main effect of salinity stress significantly affected proline concentration (Table 3.20, Table 3.23). According to the statistical data, although the proline concentration increased by approximately 6 times in medium salinity stress treated plants, it increased by 11-fold in high salinity treated plants (Table 3.23). Under medium and high salinity stress conditions, proline concentration tended to decrease in bread wheat which treated with B toxicity.

Table 3.23: Glycine betaine (GB) and proline concentrations of 4-weeks-old bread wheat (*Triticum aestivum* cv. Nusrat) and durum wheat (*Triticum durum* cv. Saricanak) grown hydroponically at different levels of salinity (control: 0 mM NaCl; medium: 60 mM NaCl; high 120 mM NaCl) and B supply (control: 2 μM; toxic: 1 mM). Data points represents the means and standard deviations of four replicates.

Cultivar	**Salinity**	**Boron**	**GB Conc. (%)**	**Proline Conc. (%)**
Nusrat	None	Control	0.9 ± 0.2	43 ± 23
		Toxic	0.9 ± 0.2	46 ± 16
	Medium	Control	1.0 ± 0.2	409 ± 291
		Toxic	1.1 ± 0.2	116 ± 63
	High	Control	1.7 ± 0.2	752 ± 696
		Toxic	1.6 ± 0.2	358 ± 354
Saricanak	None	Control	0.7 ± 0.2	48 ± 20
		Toxic	0.6 ± 0.1	34 ± 12
	Medium	Control	1.2 ± 0.2	264 ± 66
		Toxic	1.2 ± 0.2	266 ± 218
	High	Control	1.3 ± 0.1	469 ± 305
		Toxic	1.3 ± 0.2	414 ± 231

GB Conc. $HSD_{0.05}$ (S; BT; CV; SxBT; BTxCV; SxCV; SxBTxCV)= (0.1; *n.s.*; 0.1; *n.s.*; *n.s.*; 0.2; *n.s.*)
Proline Conc. $HSD_{0.05}$ (S; BT; CV; SxBT; BTxCV; SxCV; SxBTxCV)= (258; *n.s.*; *n.s.*; *n.s.*; *n.s.*; *n.s.*; *n.s.*)

Salinity stress significantly increased superoxide dismutase (SOD) total enzyme activity in both bread and durum wheat cultivars under control and toxic B conditions (Table 3.20; Table 3.24). In all conditions, SOD enzyme activity was found to be higher in Nusrat compared to Saricanak. Glutathione reductase (GR) total enzyme activity decreased with salinity stress in bread wheat, while it increased in durum wheat cultivar. Although it was observed that ascorbate peroxidase (APX) total enzyme activity was higher in Nusrat variety compared to Saricanak in control and medium salinity conditions, this effect was found to be the opposite in plants were treated with high salinity stress (Table 3.24). Catalase (CAT) total enzyme activity was increased with the high salinity stress in the cultivar of Nusrat. It was observed that the main effect of B toxicity or interactions containing B toxicity did not have a statistically significant effect on any antioxidant enzyme total activities.

Table 3.24: Effects of different levels of salinity (control: 0 mM NaCl; medium: 60 mM NaCl; high 120 mM NaCl) and B supply (control: 2 µM; toxic: 1 mM) on total antioxidant enzyme; superoxide dismutase (SOD), glutathione reductase (GR), ascorbate peroxidase (AP), and catalase (CAT) activities of 4-weeks-old bread wheat (*Triticum aestivum* cv. Nusrat) and durum wheat (*Triticum durum* cv. Saricanak) grown hydroponically under growth chamber conditions. Data points represents the means and standard deviations of four replicates.

Cultivar	Salinity	Boron	SOD Act. ($U.g^{-1}$ FW)	GR Act. (-µmol NADPH g^{-1} FW min^{-1})	APX Act. (-µmol H_2O_2 g^{-1} FW min^{-1})	CAT Act. (-mmol H_2O_2 g^{-1} FW min^{-1})
Nusrat	None	Control	133 ± 16	0.90 ± 0.08	29 ± 8	1.6 ± 0.2
		Toxic	119 ± 46	0.99 ± 0.11	33 ± 2	1.8 ± 0.3
	Medium	Control	163 ± 15	0.88 ± 0.36	39 ± 8	1.5 ± 0.5
		Toxic	149 ± 50	0.80 ± 0.15	35 ± 2	1.2 ± 0.6
	High	Control	181 ± 21	0.86 ± 0.28	31 ± 9	3.3 ± 1.2
		Toxic	188 ± 37	0.67 ± 0.17	34 ± 6	2.7 ± 1.4
Saricanak	None	Control	117 ± 20	0.51 ± 0.07	24 ± 1	2.0 ± 0.6
		Toxic	92 ± 32	0.87 ± 0.06	23 ± 4	2.1 ± 1.0
	Medium	Control	135 ± 27	0.90 ± 0.08	24 ± 6	2.3 ± 0.6
		Toxic	146 ± 30	0.97 ± 0.33	29 ± 4	2.0 ± 0.3
	High	Control	138 ± 25	0.99 ± 0.66	34 ± 3	1.6 ± 0.3
		Toxic	168 ± 20	1.04 ± 0.18	36 ± 6	2.1 ± 0.2

SOD Act. $HSD_{0.05}$ (SS; BT; CV; SSxBT; SSxCV; BTxCV; SSxBTxCV)= (26; *n.s.*; 18; *n.s.*; *n.s.*; *n.s.*; *n.s.*)
GR Act.$HSD_{0.05}$ (SS; BT; CV; SSxBT; SSxCV; BTxCV; SSxBTxCV)= (*n.s.*; *n.s.*; *n.s.*; *n.s.*; *n.s.*; 0.42; *n.s.*)
APX Act. $HSD_{0.05}$ (SS; BT; CV; SSxBT; SSxCV; BTxCV; SSxBTxCV)= (5; *n.s.*; 3; *n.s.*; *n.s.*; 8; *n.s.*)
CAT Act. $HSD_{0.05}$ (SS; BT; CV; SSxBT; SSxCV; BTxCV; SSxBTxCV)= (0.6; *n.s.*; *n.s.*; *n.s.*; 1.1; *n.s.*; *n.s.*)

Boron toxicity decreased specific (sp.) SOD activity in bread wheat cultivar in all 3 salinity treatments, while it decreased sp. SOD activity in Saricanak only in the non-saline media (Table 3.20; Table 3.25). It was determined that sp. GR activity of Nusrat was generally lower than durum wheat cultivar. Under control B conditions, specific APX enzyme activities of two wheat cultivars were generally higher than in plants grown in B toxic-media. In control and medium salinity conditions, Saricanak sp. CAT activity was detected higher compared to high salinity conditions. But it was found to be higher in Nusrat at high salinity conditions. It was realized that interactions containing B toxicity did not have a statistically significant effect on sp. antioxidant enzyme activities.

Table 3.25: Specific antioxidant enzyme; superoxide dismutase (SOD), glutathione reductase (GR), ascorbate peroxidase (AP), and catalase (CAT) activities of 4-weeks-old bread wheat (*Triticum aestivum* cv. Nusrat) and durum wheat (*Triticum durum* cv. Saricanak) grown hydroponically at different levels of salinity (control: 0 mM NaCl; medium: 60 mM NaCl; high 120 mM NaCl) and B supply (control: 2 µM; toxic: 1 mM). Data points represents the means and standard deviations of four replicates.

Cultivar	Salinity	Boron	Sp. SOD Act.	Sp. GR Act.	Sp. APX Act.	Sp. CAT Act.
			(U.mg^{-1} prt.)	(-nmol [NADPH] mg^{-1} prt.min^{-1})	(-µmol H_2O_2 mg^{-1} prt. min^{-1})	
Nusrat	None	Control	12.2 ± 2.1	84 ± 21	2.8 ± 1.2	151 ± 34
		Toxic	8.6 ± 3.3	72 ± 10	2.4 ± 0.1	130 ± 24
	Medium	Control	14.5 ± 2.1	79 ± 33	3.5 ± 0.7	133 ± 37
		Toxic	12.1 ± 4.2	61 ± 13	2.9 ± 0.6	101 ± 48
	High	Control	14.2 ± 4.0	64 ± 11	2.5 ± 0.9	244 ± 60
		Toxic	12.6 ± 1.4	45 ± 11	2.4 ± 1.0	170 ± 45
Saricanak	None	Control	15.2 ± 2.3	67 ± 7	3.1 ± 0.1	254 ± 60
		Toxic	9.8 ± 3.4	95 ± 20	2.5 ± 0.7	214 ± 62
	Medium	Control	12.7 ± 4.0	82 ± 10	2.2 ± 0.7	208 ± 57
		Toxic	13.5 ± 3.8	87 ± 23	2.7 ± 0.6	180 ± 21
	High	Control	13.2 ± 2.0	96 ± 59	3.3 ± 0.5	151 ± 23
		Toxic	13.0 ± 3.2	80 ± 21	2.8 ± 0.8	157 ± 24

SOD Act. $HSD_{0.05}$ (SS; BT; CV; SSxBT; SSxCV; BTxCV; SSxBTxCV)= (*n.s.*; 2; *n.s.*; *n.s.*; *n.s.*; *n.s.*)
GR Act.$HSD_{0.05}$ (SS; BT; CV; SSxBT; SSxCV; BTxCV; SSxBTxCV)= (*n.s.*; *n.s.*; 14; *n.s.*; *n.s.*; *n.s.*)
APX Act. $HSD_{0.05}$ (SS; BT; CV; SSxBT; SSxCV; BTxCV; SSxBTxCV)= (*n.s.*; *n.s.*; *n.s.*; *n.s.*; *n.s.*; 1; *n.s.*)
CAT Act. $HSD_{0.05}$ (SS; BT; CV; SSxBT; SSxCV; BTxCV; SSxBTxCV)= (*n.s.*; 26; 26; *n.s.*; *n.s.*; 66; *n.s.*)

3.2. Soil Experiment Results

3.2.1. First Soil Experiment Results

The first soil experiment was conducted to study the effects of the interaction of different levels of salinity stress with different levels of B toxicity on growth, development, and shoot concentrations of selected minerals in bread wheat under soil culture conditions. Moreover, it was established to decide salinity stress and B toxicity levels, which will be used in the following soil experiments.

It was observed that the main effects of moderate and severe B toxicity treatments cause to limit plant growth and height (Figure 3.14). Also, 25 days after sowing seeds, all three B toxicity treatments were caused chlorosis and necrosis in mature leaves of experimental plants beginning from leaf tips and margins. These symptoms were alleviated by increasing levels of salinity. However, enhancing salinity stress also was reduced plant growth.

Figure 3.14: Shoot growth of 25-days-old bread wheat (*Triticum aestivum* cv. Nusrat) grown in soil media at three different levels of soil salinity stress (control: 3.3 $dS.m^{-1}$ EC_e; medium: 5.8 $dS.m^{-1}$ EC_e; high: 8.3 $dS.m^{-1}$ EC_e) and B supply (adequate: 0 $mg.kg^{-1}$ H_3BO_3; mild: 10 $mg.kg^{-1}$ H_3BO_3; moderate: 20 $mg.kg^{-1}$ H_3BO_3; severe: 30 $mg.kg^{-1}$ H_3BO_3;).

Table 3.26: Two-way analysis of variance of the main effects of salinity and B supply and their interaction on germination percentage and plant height at different days after sowing (DAS) of 25-days-old bread wheat (*Triticum aestivum* cv. Nusrat) grown in soil media under growth chamber conditions.

Source of Variation	Germination Percentage		
	5 DAS	7 DAS	9 DAS
Soil Salinity (SS)	***	***	***
B Supply (BS)	*n.s.*	*	*n.s.*
SS*BS	*n.s.*	*n.s.*	*n.s.*
Source of Variation	**Plant Height**		
	7 DAS	14 DAS	21 DAS
Soil Salinity (SS)	***	***	***
B Supply (BS)	**	***	***
SS*BS	*n.s.*	*n.s.*	*n.s.*

n.s. Not significant; * 0.01 ≤ F Pr. < 0.05; ** 0.001 ≤ F Pr. < 0.01; *** F Pr. < 0.001

Germination percentage was significantly affected by soil salinity at 5, 7, and 9 days after sowing (DAS) (Table 3.26; Figure 3.15). Boron toxicity had no significant effect on germination percentage, except 7 DAS seedlings. The interaction between soil salinity and B toxicity had no significant effect on the germination percentage of wheat seedlings in any DAS.

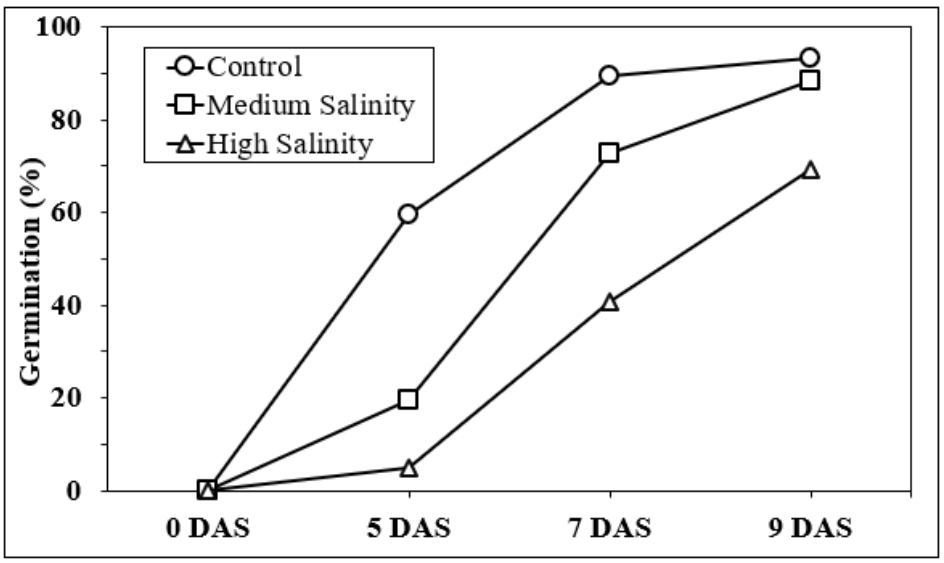

Figure 3.15: Effects of different levels of soil salinity stress (control: 3.3 $dS.m^{-1}$ EC_e; medium: 5.8 $dS.m^{-1}$ EC_e; high: 8.3 $dS.m^{-1}$ EC_e) on germination percentage of 25-days-old bread wheat (*Triticum aestivum* cv. Nusrat) grown in soil media under growth chamber conditions. Values are means of 4 independent pot replicates, each containing 15 plants.

Plant height was significantly affected by main effects of both soil salinity and B toxicity at 7, 14, and 21 DAS (Table 3.26; Table 3.27). Measurements at three different DAS showed that medium and high doses of salinity stress significantly reduced plant height. Plant height was not affected by mild B toxicity treatment. However, moderate and severe B toxicity applications significantly reduced plant height at 7, 14, and 21 DAS.

Table 3.27: Plant height of 25-days-old bread wheat (*Triticum aestivum* cv. Nusrat) grown in soil culture at different levels of soil salinity stress (control: 3.3 $dS.m^{-1}$ EC_e; medium: 5.8 $dS.m^{-1}$ EC_e; high: 8.3 $dS.m^{-1}$ EC_e) and B supply (adequate: 0 $mg.kg^{-1}$ H_3BO_3; mild: 10 $mg.kg^{-1}$ H_3BO_3; moderate: 20 $mg.kg^{-1}$ H_3BO_3; severe: 30 $mg.kg^{-1}$ H_3BO_3). Data points represents the means and standard deviations of four replicates.

Salinity	**Boron Supply**	**Plant Height ($cm.plant^{-1}$)**		
		7 DAS	**14 DAS**	**21 DAS**
Control	Adequate B	6.1 ± 1.8	22.8 ± 1.7	33.7 ± 3.0
	Mild B Tox.	6.7 ± 1.7	21.8 ± 1.6	32.2 ± 1.4
	Moderate B Tox.	5.7 ± 1.5	18.9 ± 1.8	28.9 ± 2.6
	Severe B Tox.	4.2 ± 1.2	16.4 ± 0.9	25.8 ± 0.9
Medium	Adequate B	3.5 ± 1.5	17.1 ± 2.7	27.6 ± 4.4
	Mild B Tox.	3.6 ± 1.1	17.1 ± 1.6	26.5 ± 2.3
	Moderate B Tox.	3.5 ± 1.2	16.1 ± 2.6	25.0 ± 3.5
	Severe B Tox.	2.1 ± 0.8	12.9 ± 1.7	21.6 ± 0.6
High	Adequate B	2.7 ± 0.9	13.0 ± 2.7	22.6 ± 3.7
	Mild B Tox.	2.6 ± 0.7	12.2 ± 1.8	22.2 ± 1.8
	Moderate B Tox.	1.9 ± 0.9	11.9 ± 2.9	21.0 ± 2.7
	Severe B Tox.	1.4 ± 0.8	8.9 ± 2.2	19.5 ± 2.4

Plant Height 7 DAS $HSD_{0.05}$ (SS; BS; SSxBS) = (1.1; 1.3; *n.s.*)
Plant Height 14 DAS $HSD_{0.05}$ (SS; BS; SSxBS) = (1.8; 2.3; *n.s.*)
Plant Height 21 DAS $HSD_{0.05}$ (SS; BS; SSxBS) = (2.2; 2.8; *n.s.*)

According to statistical analysis, the highest shoot fresh weight (FW) was determined in plants that were grown in control salinity conditions (Figure 3.16). Shoot FW reduced in plants that were grown in medium and high soil salinity conditions. Under control salinity, moderate and severe B toxicity treatments reduced significantly shoot FW when applied to plants.

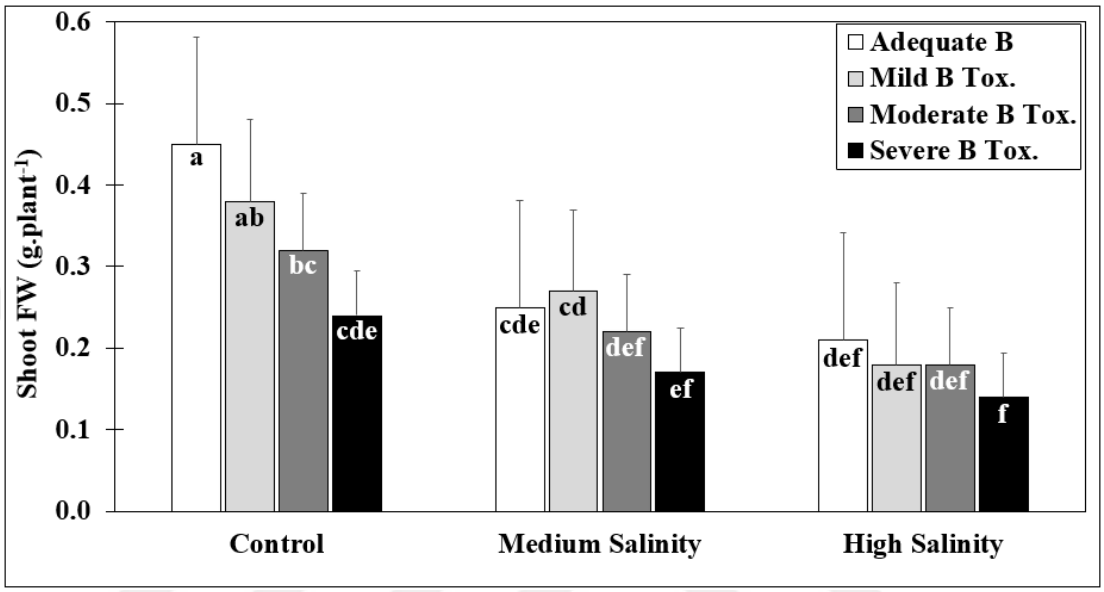

Figure 3.16: Effects of different levels of soil salinity stress (control: 3.3 $dS.m^{-1}$ EC_e; medium: 5.8 $dS.m^{-1}$ EC_e; high: 8.3 $dS.m^{-1}$ EC_e) and B supply (adequate: 0 $mg.kg^{-1}$ H_3BO_3; mild: 10 $mg.kg^{-1}$ H_3BO_3; moderate: 20 $mg.kg^{-1}$ H_3BO_3; severe: 30 $mg.kg^{-1}$ H_3BO_3) on shoot fresh weight (FW) of 25-days-old bread wheat (*Triticum aestivum* cv. Nusrat) grown in soil media under growth chamber conditions. Different letters indicate significant differences between means according to two-way ANOVA and Tukey' HSD test ($p \leq 0.05$)

Similar to the results of the shoot FW, the highest amount of shoot dry weight (DW) was found in wheat plants that are grown in non-saline conditions (Figure 3.17). The amount of DW drastically decreased in plants that were grown in medium and high salinity conditions. Under non-saline soil conditions, B toxicity treatment reduced the shoot DW. The increase of the B toxicity dose tended to decrease the amount of DW of wheat plants grown under control, medium, and high soil salinity conditions. Under the non-saline condition, severe B toxicity significantly reduced the shoot DW when compared control B condition.

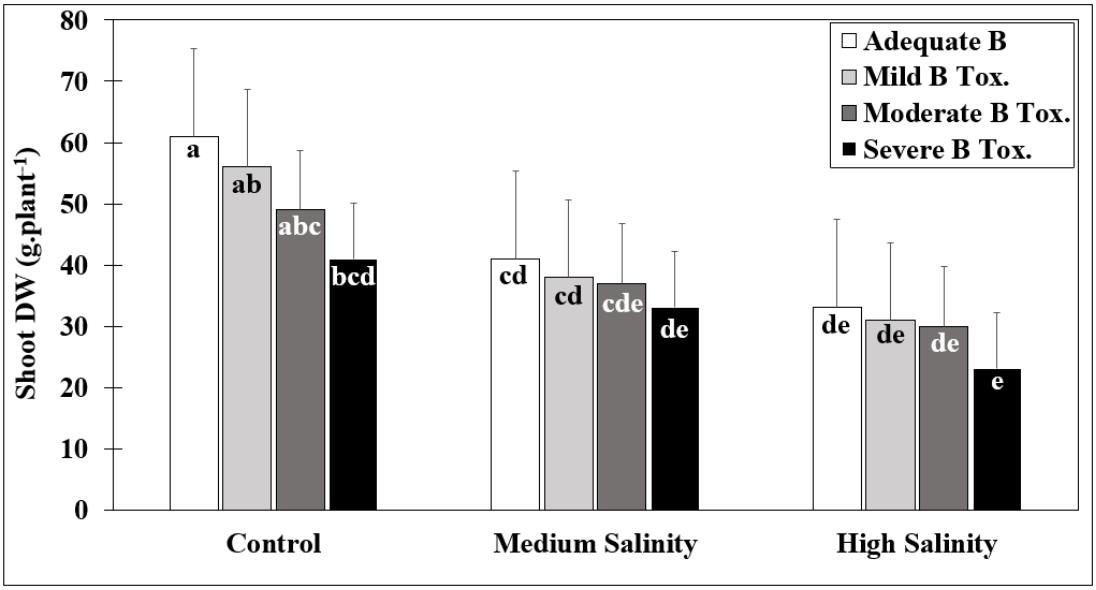

Figure 3.17: Shoot dry weight (DW) of 25-days-old bread wheat (*Triticum aestivum* cv. Nusrat) grown in soil culture at different levels of soil salinity stress (control: 3.3 $dS.m^{-1}$ EC_e; medium: 5.8 $dS.m^{-1}$ EC_e; high: 8.3 $dS.m^{-1}$ EC_e) and B supply (adequate: 0 $mg.kg^{-1}$ H_3BO_3; mild: 10 $mg.kg^{-1}$ H_3BO_3; moderate: 20 $mg.kg^{-1}$ H_3BO_3; severe: 30 $mg.kg^{-1}$ H_3BO_3). Different letters indicate significant differences between means according to two-way ANOVA and Tukey' HSD test ($p \leq 0.05$)

Medium and high salinity stress significantly reduced water content, but no difference was found between salinity doses on the effect of water content (Table 3.28). On the other hand, the application of B toxicity declined the water content gradually. It was found that the interaction of salinity stress and B toxicity treatments had a synergistic impact on water content.

Table 3.28: Effects of various levels of soil salinity stress (control: 3.3 $dS.m^{-1}$ EC_e; medium: 5.8 $dS.m^{-1}$ EC_e; high: 8.3 $dS.m^{-1}$ EC_e) and B supply (adequate: 0 $mg.kg^{-1}$ H_3BO_3; mild: 10 $mg.kg^{-1}$ H_3BO_3; moderate: 20 $mg.kg^{-1}$ H_3BO_3; severe: 30 $mg.kg^{-1}$ H_3BO_3) on water content of 25-days-old bread wheat (*Triticum aestivum* cv. Nusrat) grown in soil media under growth chamber conditions. Different letters indicate significant differences between means according to two-way ANOVA and Tukey' HSD test ($p \leq 0.05$)

Salinity	**B Supply**	**Water content**
Control	Adequate B	6.4 a
	Mild B Tox.	5.9 ab
	Moderate B Tox.	5.4 ab
	Severe B Tox.	4.9 bc
Medium	Adequate B	5.2 b
	Mild B Tox.	6.0 ab
	Moderate B Tox.	5.0 bc
	Severe B Tox.	4.1 c
High	Adequate B	5.3 b
	Mild B Tox.	4.9 bc
	Moderate B Tox.	5.0 bc
	Severe B Tox.	5.1 bc

Shoot B concentration significantly increased under mild, moderate, and severe B toxicity conditions (Table 3.29). Under toxic B circumstances, B concentration decreased in plants grown in medium and high salinity stress conditions. Shoot Na concentration significantly increased with medium and high salinity stress application. However, under the same conditions, shoot K concentration decreased.

Table 3.29: Selected mineral concentrations of 25-days-old bread wheat (*Triticum aestivum* cv. Nusrat) grown in soil culture at different levels of soil salinity stress (control: 3.3 $dS.m^{-1}$ EC_e; medium: 5.8 $dS.m^{-1}$ EC_e; high: 8.3 $dS.m^{-1}$ EC_e) and B supply (adequate: 0 $mg.kg^{-1}$ H_3BO_3; mild: 10 $mg.kg^{-1}$ H_3BO_3; moderate: 20 $mg.kg^{-1}$ H_3BO_3; severe: 30 $mg.kg^{-1}$ H_3BO_3). Different letters indicate significant differences between means according to two-way ANOVA and Tukey' HSD test ($p \leq 0.05$)

Salinity	**Boron Supply**	**Shoot Mineral Concentrations**		
		B ($mg.kg^{-1}$)	**Na (%)**	**K (%)**
Control	Adequate B	8 g	0.04 d	4.2 a
	Mild B Tox.	289 e	0.04 d	4.0 ab
	Moderate B Tox.	682 b	0.06 d	4.0 abc
	Severe B Tox.	1022 a	0.08 cd	3.8 bcde
Medium	Adequate B	26 g	0.12 cd	3.9 bcd
	Mild B Tox.	181 f	0.15 cd	3.8 bcde
	Moderate B Tox.	394 d	0.16 cd	3.7 cdef
	Severe B Tox.	658 b	0.19 bc	3.6 def
High	Adequate B	22 g	0.36 a	3.7 cdef
	Mild B Tox.	140 f	0.30 ab	3.6 def
	Moderate B Tox.	333 de	0.36 a	3.5 f
	Severe B Tox.	492 c	0.34 ab	3.7 def

3.2.2. Second Soil Experiment Results

The second soil experiment was conducted to study the effects of the interaction of different levels of salinity stress with different levels of B toxicity on growth, development, shoot accumulation of selected minerals, organic osmolyte accumulation, antioxidative enzyme activities, and oxidative damage in two bread wheat and two durum wheat cultivars under soil culture conditions.

Under the non-saline and saline conditions, moderate and severe B toxicity was caused chlorosis and necrosis in mature leaves of experimental plants beginning from leaf tips and margins (Figure 3.19). This symptom was alleviated by increasing levels of salinity. However, increasing salinity also was reduced plant growth under moderate and severe B toxicity. Similar to salinity, to the main factor of the B toxicity also decreased plant growth. But when compared the non-saline and saline conditions, the primary factor of reduction of plant growth was seen as salinity stress. Besides, plant growth reductions between cultivars were observed differently. In comparison to other wheat cultivars, Fuatbey-2000, a durum wheat cultivar, was more affected by salinity stress and B toxicity (Figure 3.18).

Figure 3.18: Shoot growth of 40-days-old cultivars of bread wheat (*Triticum aestivum* cv. Nusrat and Adana-99) and durum wheat (*Triticum durum* cv. Saricanak and Fuatbey-2000) grown in soil at three different levels of soil salinity stress (control: 3.5 $dS.m^{-1}$ EC_e; medium: 6.0 $dS.m^{-1}$ EC_e; high: 8.5 $dS.m^{-1}$ EC_e) and B supply (adequate: 0 $mg.kg^{-1}$ H_3BO_3; moderate: 15 $mg.kg^{-1}$ H_3BO_3; severe: 30 $mg.kg^{-1}$ H_3BO_3).

In this second soil experiment, ANOVA revealed highly significant main effects of soil salinity and cultivar and their interaction on the germination percentage of 8, 12, and 14 DAS bread and durum wheat plants grown under greenhouse conditions (Table 3.30). Medium soil salinity did not affect the germination percentage at 14 DAS. But high soil salinity treatment significantly reduced the germination percentage at 14 DAS (Table 3.31).

In the germination percentage results was collected at 14 DAS, the highest germination percentage was found in the Adana-99 cultivar, which is a bread wheat species. No significant difference was found in terms of 14 DAS germination percentage results between the other bread cultivar Nusrat, and the two durum varieties: Saricanak and Fuatbey-2000 (Table 3.30; Table 3.31).

While the negative effect of B toxicity on germination percentage was statistically significant in 8 and 12 DAS, this effect was lost in 14 DAS (Table 3.30; Table 3.31). The effect of soil salinity*B toxicity interaction on the germination percentage is statistically significant in 12 and 14 DAS (Table 3.30). Because the salinity stress and B toxicity interaction displayed an additive effect on germination percentage at 14 DAS, germination percentage decreased with increasing stress doses, especially in Fuatbey-2000 of durum wheat cultivar (Table 3.31).

Table 3.30: Analysis of variance of the main effects of salinity, B supply, and cultivar and their interactions on germination percentage (8, 12, and 14 DAS) and shoot dry weight (DW) of 40-days-old cultivars of bread wheat (*Triticum aestivum* cv. Nusrat and Adana-99) and durum wheat (*Triticum durum* cv. Saricanak and Fuatbey-2000) grown in soil under greenhouse conditions.

Source of Variation	**Germination Percentage (8 DAS)**	**Germination Percentage (12 DAS)**	**Germination Percentage (14 DAS)**	**Shoot DW**
Soil Salinity (SS)	***	***	***	***
B Supply (BS)	**	*	*n.s.*	***
Cultivar (CV)	***	**	***	*
SS*BS	*n.s.*	**	**	*n.s.*
BS*CV	*n.s.*	*n.s.*	*	*n.s.*
SS*CV	*	***	***	*n.s.*
SS*BS*CV	*n.s.*	*n.s.*	*n.s.*	*n.s.*

n.s. Not significant; * $0.01 \leq$ F Pr. < 0.05; ** $0.001 \leq$ F Pr. < 0.01; *** F Pr. < 0.001

The main effects of salinity stress, B toxicity and, cultivar on shoot DW showed significant effects in bread and durum wheat plants grown under greenhouse conditions (Table 3.30). Any interaction situation had no statistically significant effect on the wheat shoot DW.

Medium and high soil salinity treatments significantly reduced shoot DW (Table 3.31). Shoot DW was decreased significantly in the presence of moderate and severe toxic-B. While the highest shoot DW was determined in Adana-99, the lowest DW amount was found in Fuatbey-2000. It was assessed that the Nusrat and Saricanak shoot DW were statistically similar.

Table 3.31: Effects of different doses of soil salinity stress (control: 3.5 $dS.m^{-1}$ EC_e; medium: 6.0 $dS.m^{-1}$ EC_e; high: 8.5 $dS.m^{-1}$ EC_e) and B supply (adequate: 0 $mg.kg^{-1}$ H_3BO_3; moderate: 15 $mg.kg^{-1}$ H_3BO_3; severe: 30 $mg.kg^{-1}$ H_3BO_3) on germination percentage of 14 DAS and shoot dry weight (DW) of 40-days-old cultivars of bread wheat (*Triticum aestivum* cv. Nusrat and Adana-99) and durum wheat (*Triticum durum* cv. Saricanak and Fuatbey-2000) grown in soil under greenhouse conditions. Data points represents the means and standard deviations of four replicates.

B Supply	Soil Salinity	Cultivar	Germination (%)	Shoot DW ($g.plant^{-1}$)
Adequate B	Control	Nusrat	88 ± 6	0.53 ± 0.14
		Adana-99	100 ± 0	0.64 ± 0.14
		Saricanak	88 ± 3	0.54 ± 0.19
		Fuatbey-2000	100 ± 0	0.45 ± 0.10
	Medium	Nusrat	92 ± 6	0.39 ± 0.05
		Adana-99	97 ± 4	0.41 ± 0.03
		Saricanak	90 ± 7	0.33 ± 0.06
		Fuatbey-2000	97 ± 4	0.32 ± 0.07
	High	Nusrat	88 ± 6	0.33 ± 0.13
		Adana-99	95 ± 6	0.42 ± 0.03
		Saricanak	87 ± 8	0.30 ± 0.14
		Fuatbey-2000	87 ± 5	0.26 ± 0.10
Moderate B Toxicity	Control	Nusrat	93 ± 5	0.45 ± 0.09
		Adana-99	98 ± 3	0.59 ± 0.11
		Saricanak	88 ± 3	0.40 ± 0.18
		Fuatbey-2000	93 ± 9	0.38 ± 0.03
	Medium	Nusrat	97 ± 4	0.29 ± 0.04
		Adana-99	97 ± 4	0.31 ± 0.05
		Saricanak	88 ± 6	0.41 ± 0.09
		Fuatbey-2000	97 ± 4	0.38 ± 0.18
	High	Nusrat	92 ± 6	0.34 ± 0.06
		Adana-99	100 ± 0	0.25 ± 0.04
		Saricanak	92 ± 10	0.28 ± 0.03
		Fuatbey-2000	82 ± 21	0.19 ± 0.08
Severe B Toxicity	Control	Nusrat	92 ± 3	0.39 ± 0.08
		Adana-99	100 ± 0	0.26 ± 0.07
		Saricanak	98 ± 3	0.35 ± 0.09
		Fuatbey-2000	98 ± 3	0.32 ± 0.06
	Medium	Nusrat	92 ± 6	0.19 ± 0.04
		Adana-99	100 ± 0	0.21 ± 0.07
		Saricanak	93 ± 8	0.29 ± 0.10
		Fuatbey-2000	92 ± 3	0.12 ± 0.04
	High	Nusrat	85 ± 11	0.14 ± 0.08
		Adana-99	93 ± 5	0.09 ± 0.04
		Saricanak	82 ± 17	0.13 ± 0.04
		Fuatbey-2000	55 ± 24	0.12 ± 0.04

Germination 14 DAS $HSD_{0.05}$ (SS; BS; CV; SSxBS; SSxCV; BSxCV; SSxBSxCV)= (4; *n.s.*; 5; 9; 11; 11; *n.s.*)
Shoot DW $HSD_{0.05}$ (SS; BS; CV; SSxBS; SSxCV; BSxCV; SSxBSxCV)= (0.05; 0.05; 0.06; *n.s.*; *n.s.*; *n.s.*; *n.s.*)

Analysis of variance revealed significant impacts of main effects of salinity, B toxicity, and cultivar and two-way interactions of these stress factors on the 40 DAS shoot B concentration of bread and durum wheat species grown under greenhouse conditions (Table 3.32). Shoot B content ANOVA results are also very similar to the B concentration. However, while salinity*cultivar interaction had no significant effect on shoot B content, 3-way interaction of salinity, B toxicity, and cultivar significantly affected shoot B content (Table 3.32).

Table 3.32: Analysis of variance of the main effects of salinity, B supply, and cultivar and their interactions on concentrations and contents of B and Na of 40-days-old cultivars of bread wheat (*Triticum aestivum* cv. Nusrat and Adana-99) and durum wheat (*Triticum durum* cv. Saricanak and Fuatbey-2000) grown in soil under greenhouse conditions.

Source of Variation	Shoot B Concentration	Shoot B Content	Shoot Na Concentration	Shoot Na Content
Soil Salinity (SS)	***	***	***	***
B Supply (BS)	***	***	*n.s.*	***
Cultivar (CV)	***	**	***	***
SS*BS	***	***	*n.s.*	**
BS*CV	***	**	*n.s.*	**
SS*CV	**	*n.s.*	***	***
SS*BS*CV	*n.s.*	*	*	*n.s.*

n.s. Not significant; * 0.01 ≤ F Pr. < 0.05; ** 0.001 ≤ F Pr. < 0.01; *** F Pr. < 0.001

Table 3.33: Shoot B accumulation of 40-days-old bread wheat (*Triticum aestivum* cv. Nusrat and Adana-99) and durum wheat (*Triticum durum* cv. Saricanak and Fuatbey-2000) grown in soil culture at different levels of soil salinity stress (control: 3.5 $dS.m^{-1}$ EC_e; medium: 6.0 $dS.m^{-1}$ EC_e; high: 8.5 $dS.m^{-1}$ EC_e) and B supply (adequate: 0 $mg.kg^{-1}$ H_3BO_3; moderate: 15 $mg.kg^{-1}$ H_3BO_3; severe: 30 $mg.kg^{-1}$ H_3BO_3). Data points represents the means and standard deviations of four replicates.

B Supply	Soil Salinity	Cultivar	Shoot B Conc. ($mg.kg^{-1}$)	Shoot B Content ($\mu g.plant^{-1}$)
Adequate B	Control	Nusrat	11 ± 1	6 ± 1
		Adana-99	11 ± 2	7 ± 1
		Saricanak	13 ± 1	7 ± 2
		Fuatbey-2000	13 ± 3	6 ± 0
	Medium	Nusrat	12 ± 3	5 ± 1
		Adana-99	12 ± 1	5 ± 1
		Saricanak	13 ± 1	4 ± 0
		Fuatbey-2000	15 ± 2	5 ± 1
	High	Nusrat	12 ± 2	4 ± 1
		Adana-99	11 ± 0	5 ± 0
		Saricanak	12 ± 2	4 ± 2
		Fuatbey-2000	13 ± 2	3 ± 1
Moderate B Toxicity	Control	Nusrat	332 ± 70	150 ± 36
		Adana-99	409 ± 63	245 ± 85
		Saricanak	517 ± 68	210 ± 106
		Fuatbey-2000	486 ± 26	184 ± 22
	Medium	Nusrat	347 ± 33	99 ± 5
		Adana-99	327 ± 66	98 ± 8
		Saricanak	349 ± 30	143 ± 29
		Fuatbey-2000	398 ± 67	143 ± 55
	High	Nusrat	307 ± 19	103 ± 13
		Adana-99	311 ± 70	76 ± 7
		Saricanak	329 ± 19	93 ± 12
		Fuatbey-2000	394 ± 61	77 ± 36
Severe B Toxicity	Control	Nusrat	990 ± 161	381 ± 93
		Adana-99	1186 ± 101	310 ± 76
		Saricanak	1337 ± 67	472 ± 119
		Fuatbey-2000	1349 ± 185	424 ± 88
	Medium	Nusrat	1007 ± 90	176 ± 20
		Adana-99	991 ± 157	199 ± 66
		Saricanak	1171 ± 144	336 ± 108
		Fuatbey-2000	1140 ± 88	142 ± 53
	High	Nusrat	712 ± 74	104 ± 69
		Adana-99	670 ± 55	59 ± 26
		Saricanak	733 ± 126	94 ± 35
		Fuatbey-2000	902 ± 58	107 ± 33

Shoot B Conc. $HSD_{0.05}$ (SS; BS; CV; SSxBS; SSxCV; BSxCV; SSxBSxCV)= (37; 37; 46; 84; 105; 105; *n.s.*)
Shoot B Cont. $HSD_{0.05}$ (SS; BS; CV; SSxBS; SSxCV; BSxCV; SSxBSxCV)= (25; 25; 32; 58; *n.s.;* 71; 141)

Moderate and severe B toxicity applications significantly increased shoot B accumulation (Table 3.32; Table 3.33). Boron concentration and content, which increased with any application of toxic-B to plants, significantly decreased in the presence of soil salinity. These reductions of B accumulation were exacerbated with the enhancing salinity stress (Table 3.33; Figure 3.19; Figure 3.20).

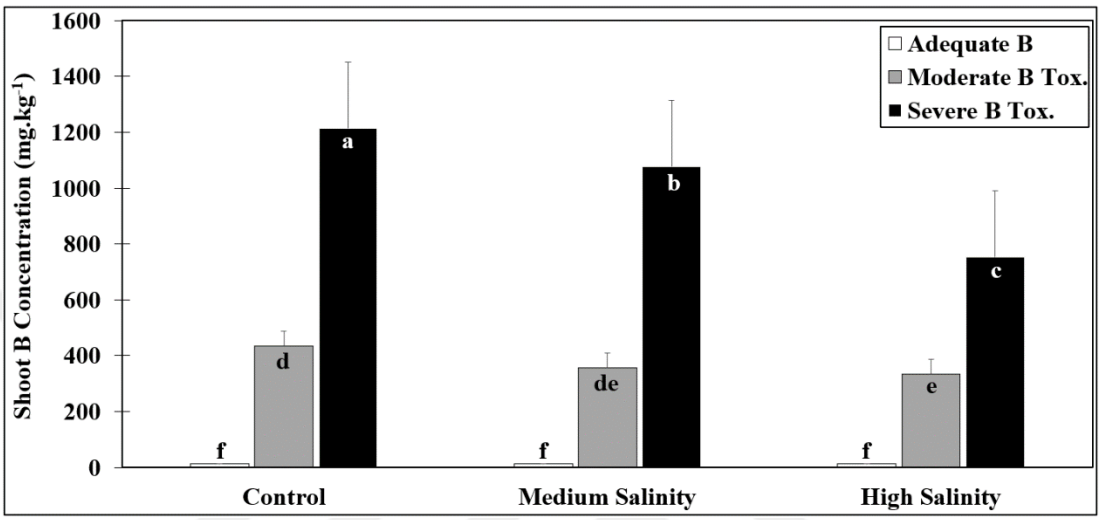

Figure 3.19: Shoot B concentrations of 40-days-old wheat grown in soil culture at different levels of soil salinity stress (control: 3.5 $dS.m^{-1}$ EC_e; medium: 6.0 $dS.m^{-1}$ EC_e; high: 8.5 $dS.m^{-1}$ EC_e) and B supply (adequate: 0 $mg.kg^{-1}$ H_3BO_3; moderate: 15 $mg.kg^{-1}$ H_3BO_3; severe: 30 $mg.kg^{-1}$ H_3BO_3) under greenhouse conditions. Different letters indicate significant differences between means according to two-way ANOVA and Tukey's HSD test ($p \leq 0.05$).

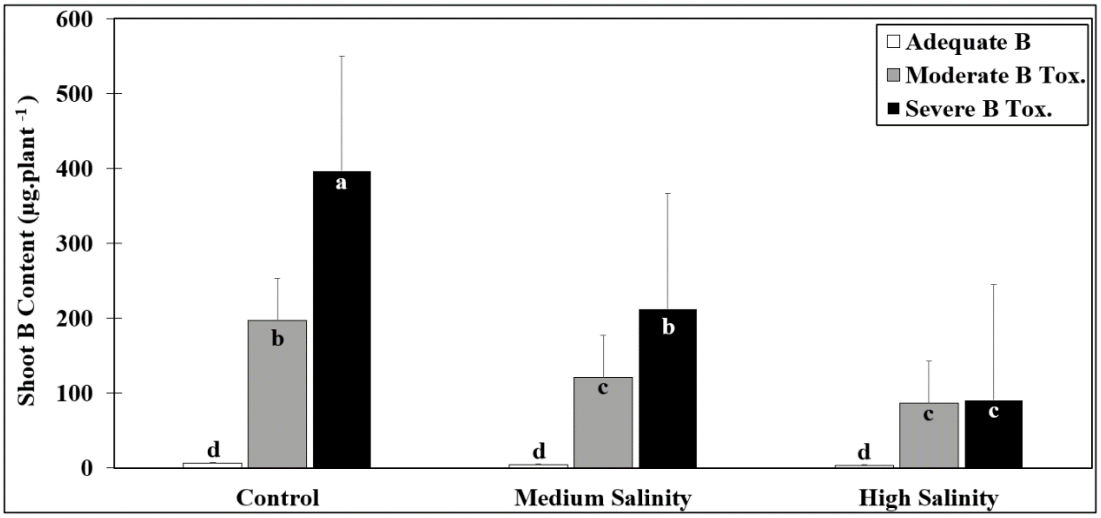

Figure 3.20: Shoot B content of 40-days-old wheat grown in soil culture at different levels of soil salinity stress (control: 3.5 $dS.m^{-1}$ EC_e; medium: 6.0 $dS.m^{-1}$ EC_e; high: 8.5 $dS.m^{-1}$ EC_e) and B supply (adequate: 0 $mg.kg^{-1}$ H_3BO_3; moderate: 15 $mg.kg^{-1}$ H_3BO_3; severe: 30 $mg.kg^{-1}$ H_3BO_3). Different letters indicate significant differences between means according to two-way ANOVA and Tukey's HSD test ($p \leq 0.05$).

Boron accumulation was not found statistically significant between cultivars treated with adequate and moderate B toxicity (Figure 3.21; Figure 3.22). However, when severe B toxicity applied to plants, shoot B concentration were found to be higher in durum wheat cultivars (Saricanak and Fuatbey-2000) when compared to bread wheat cultivars (Nusrat and Adana-99) (Figure 3.22). Also, it was determined that shoot B content significantly higher in the Saricanak durum wheat cultivar than other wheat varieties (Figure 3.23).

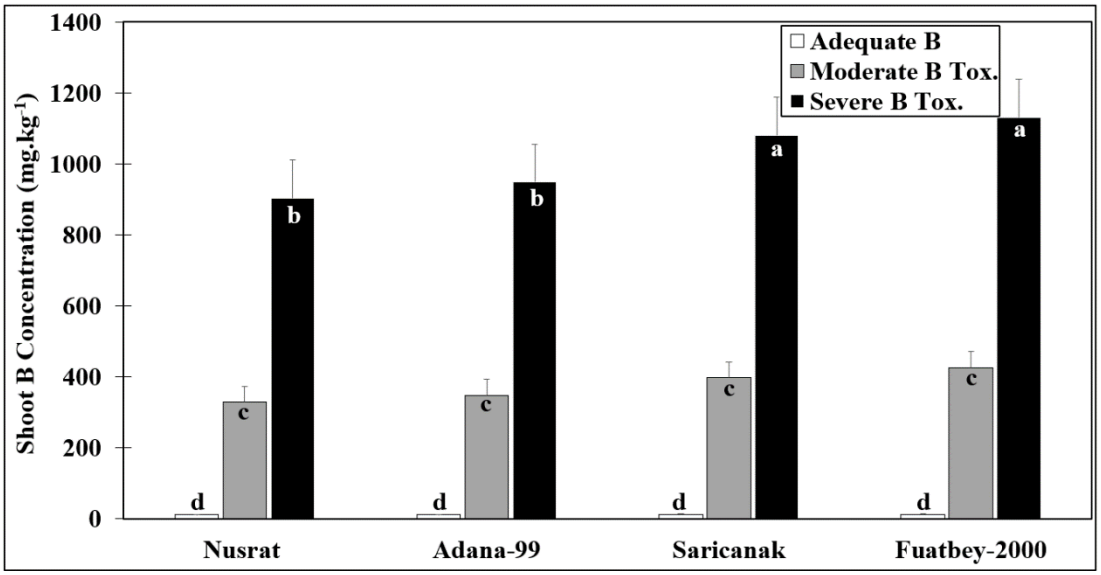

Figure 3.21: Effects of different doses of B supply (adequate: 0 mg.kg^{-1} H_3BO_3; moderate: 15 mg.kg^{-1} H_3BO_3; severe: 30 mg.kg^{-1} H_3BO_3) on shoot B concentrations of 40-days-old cultivars of bread wheat (*Triticum aestivum* cv. Nusrat and Adana-99) and durum wheat (*Triticum durum* cv. Saricanak and Fuatbey-2000) grown in soil under greenhouse conditions. Different letters indicate significant differences between means according to two-way ANOVA and Tukey's HSD test ($p \leq 0.05$).

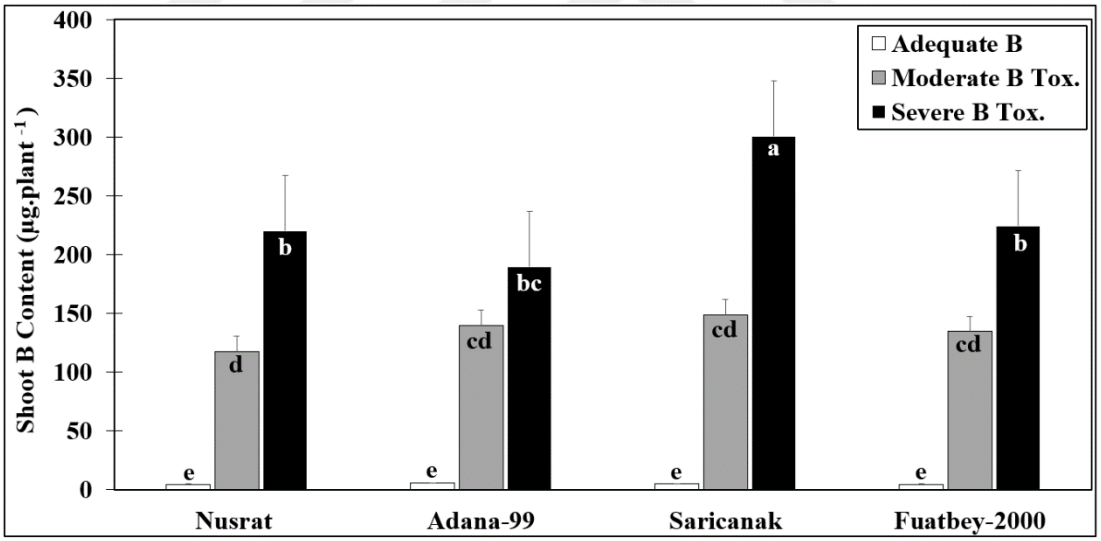

Figure 3.22: Effects of different doses of B supply (adequate: 0 mg.kg^{-1} H_3BO_3; moderate: 15 mg.kg^{-1} H_3BO_3; severe: 30 mg.kg^{-1} H_3BO_3) on shoot B contents of 40-days-old cultivars of bread wheat (*Triticum aestivum* cv. Nusrat and Adana-99) and durum wheat (*Triticum durum* cv. Saricanak and Fuatbey-2000) grown in soil under greenhouse conditions. Different letters indicate significant differences between means according to two-way ANOVA and Tukey's HSD test ($p \leq 0.05$).

Medium and high soil salinity stress treatments significantly increased shoot Na concentration and content (Table 3.32; Table 3.34). Boron toxicity had no significant effect on Na concentration. The highest Na concentration was detected in the durum wheat varieties, Fuatbey-2000 and Saricanak. It was determined that the Na concentration in durum wheat varieties was 5-fold higher than the bread wheat varieties Nusrat and Adana-99 (Table 3.32; Table 3.34).

Moderate B toxicity application did not significantly affect shoot Na content (Table 3.32; Table 3.34). However, shoot Na content decreased to approximately half in plants treated with severe B toxicity compared to the B-adequate plants. The shoot Na content of durum wheat cultivars are 6-times higher than bread wheat varieties.

Table 3.34: Effects of different doses of soil salinity stress (control: 3.5 $dS.m^{-1}$ EC_e; medium: 6.0 $dS.m^{-1}$ EC_e; high: 8.5 $dS.m^{-1}$ EC_e) and B supply (adequate: 0 $mg.kg^{-1}$ H_3BO_3; moderate: 15 $mg.kg^{-1}$ H_3BO_3; severe: 30 $mg.kg^{-1}$ H_3BO_3) on shoot Na accumulation of 40-days-old cultivars of bread wheat (*Triticum aestivum* cv. Nusrat and Adana-99) and durum wheat (*Triticum durum* cv. Saricanak and Fuatbey-2000) grown in soil under greenhouse conditions. Data points represents the means and standard deviations of four replicates.

B Supply	Soil Salinity	Cultivar	Shoot Na Conc. (%)	Shoot Na Content (mg. $plant^{-1}$)
Adequate B	Control	Nusrat	0.01 ± 0.01	0.0 ± 0.1
		Adana-99	0.01 ± 0.00	0.1 ± 0.0
		Saricanak	0.40 ± 0.41	1.8 ± 1.1
		Fuatbey-2000	0.23 ± 0.01	1.0 ± 0.2
	Medium	Nusrat	0.09 ± 0.08	0.3 ± 0.4
		Adana-99	0.11 ± 0.05	0.5 ± 0.2
		Saricanak	0.89 ± 0.07	2.9 ± 0.6
		Fuatbey-2000	1.20 ± 0.10	3.8 ± 0.9
	High	Nusrat	0.32 ± 0.24	0.8 ± 0.4
		Adana-99	0.22 ± 0.05	1.0 ± 0.3
		Saricanak	1.40 ± 0.10	4.2 ± 1.9
		Fuatbey-2000	1.78 ± 0.10	4.6 ± 1.5
Moderate B Toxicity	Control	Nusrat	0.00 ± 0.00	0.0 ± 0.0
		Adana-99	0.01 ± 0.00	0.0 ± 0.0
		Saricanak	0.16 ± 0.02	0.6 ± 0.2
		Fuatbey-2000	0.20 ± 0.01	0.7 ± 0.0
	Medium	Nusrat	0.10 ± 0.05	0.3 ± 0.1
		Adana-99	0.16 ± 0.10	0.4 ± 0.2
		Saricanak	0.87 ± 0.07	3.6 ± 0.8
		Fuatbey-2000	1.04 ± 0.06	3.8 ± 1.7
	High	Nusrat	0.43 ± 0.06	1.5 ± 0.4
		Adana-99	0.34 ± 0.14	0.8 ± 0.3
		Saricanak	1.25 ± 0.21	3.6 ± 1.0
		Fuatbey-2000	1.93 ± 0.29	3.8 ± 1.8
Severe B Toxicity	Control	Nusrat	0.00 ± 0.00	0.0 ± 0.0
		Adana-99	0.01 ± 0.01	0.0 ± 0.0
		Saricanak	0.17 ± 0.02	0.6 ± 0.1
		Fuatbey-2000	0.18 ± 0.03	0.5 ± 0.1
	Medium	Nusrat	0.14 ± 0.07	0.2 ± 0.1
		Adana-99	0.17 ± 0.07	0.4 ± 0.3
		Saricanak	0.84 ± 0.22	2.6 ± 1.2
		Fuatbey-2000	0.99 ± 0.12	1.2 ± 0.5
	High	Nusrat	0.16 ± 0.08	0.2 ± 0.1
		Adana-99	0.35 ± 0.05	0.3 ± 0.1
		Saricanak	1.55 ± 0.08	1.9 ± 0.5
		Fuatbey-2000	1.86 ± 0.21	2.2 ± 0.5

Shoot Na Conc. $HSD_{0.05}$ (SS; BS; CV; SSxBS; SSxCV; BSxCV; SSxBSxCV)= (0.06; *n.s.*; 0.07; *n.s.*; 0.2; *n.s.*; 0.35).
Shoot Na Cont. $HSD_{0.05}$ (SS; BS; CV; SSxBS; SSxCV; BSxCV; SSxBSxCV)= (0.4; 0.4; 0.5; 0.8; 1; 1; *n.s.*).

Shoot K concentration was decreased significantly under medium and high soil salinity stress. Also, shoot K concentration differed between cultivars and species. Bread wheat cultivars had higher shoot K concentrations than durum wheat cultivars (Table 3.35; Table 3.36).

Table 3.35: Analysis of variance of the main effects of salinity, B supply, and cultivar and their interactions on shoot concentrations of selected macronutrients and micronutrients of 40-days-old cultivars of bread wheat (*Triticum aestivum* cv. Nusrat and Adana-99) and durum wheat (*Triticum durum* cv. Saricanak and Fuatbey-2000) grown in soil under greenhouse conditions.

Source of Variation	**Shoot Macronutrient Concentrations**			
	K	**Ca**	**P**	**Mg**
Soil Salinity (SS)	***	*	*n.s.*	**
B Supply (BS)	***	*n.s.*	***	***
Cultivar (CV)	***	***	**	***
SS*BS	**	*n.s.*	*n.s.*	*n.s.*
BS*CV	*n.s.*	*n.s.*	*n.s.*	*n.s.*
SS*CV	***	*n.s.*	*	*n.s.*
SS*BS*CV	*	*n.s.*	*n.s.*	*n.s.*
Source of Variation	**Shoot Micronutrient Concentrations**			
	Fe	**Zn**	**Mn**	**Cu**
Soil Salinity (SS)	***	*	***	*n.s.*
B Supply (BS)	**	***	*n.s.*	***
Cultivar (CV)	***	***	***	***
SS*BS	**	*n.s.*	*n.s.*	*n.s.*
BS*CV	*n.s.*	*n.s.*	*n.s.*	*
SS*CV	**	*	*n.s.*	*n.s.*
SS*BS*CV	**	*	*n.s.*	*n.s.*

n.s. Not significant; * $0.01 \leq$ F Pr. < 0.05; ** $0.001 \leq$ F Pr. < 0.01; *** F Pr. < 0.001

High salinity stress treatment reduced shoot Ca concentration by 10%. It was determined that the shoot Ca concentration of cultivars of durum wheat was higher than the bread wheat (Table 3.35; Table 3.36). Severe B toxicity treatment significantly enhanced shoot P concentration, while moderate B toxicity application tended to decrease it (Table 3.35; Table 3.36). Whereas severe B toxicity treatment enhanced shoot P concentration significantly, moderate B toxicity application tended to decrease it. In addition to those macronutrients, shoot Mg concentration were decreased significantly by B toxicity treatments, but were increased with salinity applications. Also, bread wheat Mg concentrations were found to be higher than durum wheat varieties (Table 3.35; Table 3.36).

Table 3.36: Selected macronutrient concentrations of 40-days-old bread wheat (*Triticum aestivum* cv. Nusrat and Adana-99) and durum wheat (*Triticum durum* cv. Saricanak and Fuatbey-2000) grown in soil culture at various levels of soil salinity stress (control: 3.5 $dS.m^{-1}$ EC_e; medium: 6.0 $dS.m^{-1}$ EC_e; high: 8.5 $dS.m^{-1}$ EC_e) and B supply (adequate: 0 $mg.kg^{-1}$ H_3BO_3; moderate: 15 $mg.kg^{-1}$ H_3BO_3; severe: 30 $mg.kg^{-1}$ H_3BO_3). Data points represents the means and standard deviations of four replicates.

B Supply	Soil Salinity	Cultivar	Shoot K Conc. (%)	Shoot Ca Conc. (%)	Shoot P Conc. (%)	Shoot Mg Conc. (%)
Adequate B	Control	Nusrat	4.9 ± 0.1	0.8 ± 0.0	0.29 ± 0.03	0.201 ± 0.008
		Adana-99	4.6 ± 0.2	0.8 ± 0.1	0.33 ± 0.03	0.171 ± 0.010
		Saricanak	4.4 ± 0.3	1.2 ± 0.1	0.24 ± 0.02	0.146 ± 0.005
		Fuatbey-00	4.2 ± 0.1	1.4 ± 0.3	0.26 ± 0.05	0.149 ± 0.008
	Medium	Nusrat	5.0 ± 0.1	0.8 ± 0.1	0.29 ± 0.04	0.191 ± 0.009
		Adana-99	4.7 ± 0.2	0.8 ± 0.1	0.29 ± 0.04	0.180 ± 0.001
		Saricanak	3.9 ± 0.2	1.1 ± 0.2	0.22 ± 0.06	0.148 ± 0.018
		Fuatbey-00	3.6 ± 0.2	1.3 ± 0.3	0.27 ± 0.03	0.143 ± 0.011
	High	Nusrat	4.6 ± 0.3	1.1 ± 0.4	0.25 ± 0.03	0.197 ± 0.012
		Adana-99	4.6 ± 0.1	0.7 ± 0.1	0.26 ± 0.04	0.192 ± 0.013
		Saricanak	3.4 ± 0.2	1.1 ± 0.1	0.27 ± 0.06	0.155 ± 0.007
		Fuatbey-00	3.1 ± 0.2	1.4 ± 0.2	0.28 ± 0.02	0.148 ± 0.007
Moderate B Toxicity	Control	Nusrat	5.1 ± 0.2	0.7 ± 0.0	0.31 ± 0.03	0.173 ± 0.021
		Adana-99	4.4 ± 0.4	0.7 ± 0.1	0.27 ± 0.04	0.158 ± 0.013
		Saricanak	4.1 ± 0.6	1.1 ± 0.2	0.22 ± 0.05	0.136 ± 0.009
		Fuatbey-00	4.0 ± 0.3	1.2 ± 0.1	0.24 ± 0.04	0.140 ± 0.012
	Medium	Nusrat	4.6 ± 0.2	1.0 ± 0.2	0.24 ± 0.03	0.187 ± 0.006
		Adana-99	4.4 ± 0.2	0.9 ± 0.2	0.25 ± 0.03	0.175 ± 0.014
		Saricanak	3.8 ± 0.1	1.0 ± 0.2	0.25 ± 0.03	0.141 ± 0.013
		Fuatbey-00	4.0 ± 0.4	1.2 ± 0.3	0.27 ± 0.02	0.134 ± 0.010
	High	Nusrat	4.4 ± 0.3	1.1 ± 0.1	0.30 ± 0.05	0.178 ± 0.015
		Adana-99	4.1 ± 0.3	0.8 ± 0.1	0.21 ± 0.03	0.155 ± 0.006
		Saricanak	3.4 ± 0.4	1.2 ± 0.2	0.22 ± 0.04	0.146 ± 0.016
		Fuatbey-00	2.9 ± 0.2	1.4 ± 0.3	0.28 ± 0.03	0.152 ± 0.017
Severe B Toxicity	Control	Nusrat	5.3 ± 0.2	0.8 ± 0.0	0.31 ± 0.04	0.160 ± 0.009
		Adana-99	4.8 ± 0.1	0.9 ± 0.1	0.29 ± 0.05	0.151 ± 0.007
		Saricanak	4.9 ± 0.2	1.0 ± 0.0	0.28 ± 0.03	0.132 ± 0.008
		Fuatbey-00	4.5 ± 0.2	1.0 ± 0.1	0.30 ± 0.03	0.124 ± 0.010
	Medium	Nusrat	4.5 ± 0.1	1.0 ± 0.1	0.29 ± 0.02	0.183 ± 0.018
		Adana-99	4.8 ± 0.1	0.8 ± 0.2	0.30 ± 0.02	0.166 ± 0.008
		Saricanak	4.2 ± 0.2	1.0 ± 0.1	0.32 ± 0.07	0.141 ± 0.015
		Fuatbey-00	3.6 ± 0.1	1.2 ± 0.0	0.28 ± 0.03	0.127 ± 0.007
	High	Nusrat	4.7 ± 0.2	0.9 ± 0.1	0.29 ± 0.02	0.178 ± 0.016
		Adana-99	4.5 ± 0.2	0.9 ± 0.1	0.30 ± 0.03	0.167 ± 0.012
		Saricanak	3.4 ± 0.3	1.1 ± 0.2	0.27 ± 0.04	0.144 ± 0.012
		Fuatbey-00	3.0 ± 0.3	1.1 ± 0.2	0.33 ± 0.03	0.133 ± 0.010

Shoot K Conc. $HSD_{0.05}$ (SS; BS; CV; SSxBS; SSxCV; BSxCV; SSxBSxCV)= (0.1; 0.1; 0.1; 0.3; 0.3; *n.s.*; 0.7)
Shoot Ca Conc. $HSD_{0.05}$ (SS; BS; CV; SSxBS; SSxCV; BSxCV; SSxBSxCV)= (0.1; *n.s.*; 0.1; *n.s.*; *n.s.*; *n.s.*; *n.s.*)
Shoot P Conc. $HSD_{0.05}$ (SS; BS; CV; SSxBS; SSxCV; BSxCV; SSxBSxCV)= (*n.s.*; 0.02; 0.02; *n.s.*; 0.05; *n.s.*; *n.s.*)
Shoot Mg Conc. $HSD_{0.05}$ (SS; BS; CV; SSxBS; SSxCV; BSxCV; SSxBSxCV)= (0.005; 0.005; 0.007; *n.s.*; *n.s.*; *n.s.*; *n.s.*)

Severe B toxicity treatment did not show a positive effect on the wheat shoot K/Na ratio except for the high-saline Nusrat cultivar (Figure 3.24). Also, as expected, soil salinity application significantly decreased the shoot K/Na ratio. However, it was determined that the Nusrat and Adana-99 cultivars had a higher K/Na ratio than the durum wheat varieties (Figure 3.23).

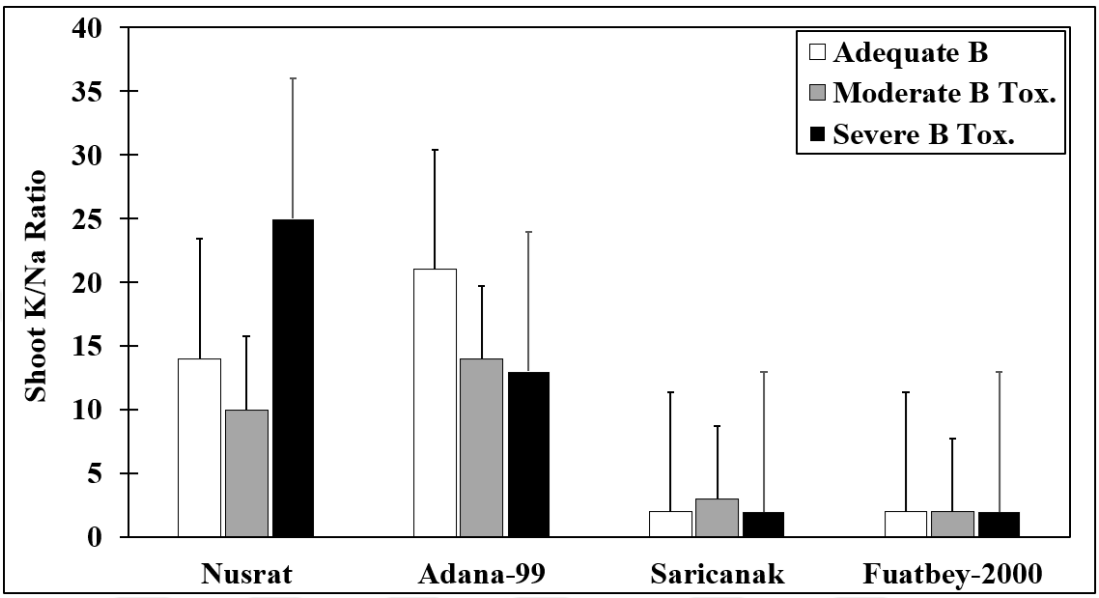

Figure 3.23: Effects of B supply (adequate: 0 $mg.kg^{-1}$ H_3BO_3; moderate: 15 $mg.kg^{-1}$ H_3BO_3; severe: 30 $mg.kg^{-1}$ H_3BO_3) on shoot K/Na ratio of 40-days-old cultivars of bread wheat (*Triticum aestivum* cv. Nusrat and Adana-99) and durum wheat (*Triticum durum* cv. Saricanak and Fuatbey-2000) grown in high saline-soil (high: 8.5 $dS.m^{-1}$ EC_e) under greenhouse conditions. Bars represents standard deviations.

Salinity treatment significantly reduced shoot Fe concentration in wheat plants (Table 3.35; Table 3.37). While moderate B toxicity application significantly increased the shoot Fe concentration, severe B toxicity treatment did not affect it. It was determined that the shoot Fe concentration of bread wheat cultivars were higher than that of durum wheat varieties (Table 3.37).

Although severe B toxicity treatment reduced the concentration of shoot Zn by 10%, moderate B toxicity decreased shoot Zn by 24% (Table 3.35; Table 3.37). Shoot Zn concentration was increased significantly as a result of medium salinity application to the wheat, but high salinity treatment did not affect it importantly (Table 3.37).

Shoot Mn concentration increased as a result of medium and high salinity application (Table 3.35; Table 3.37). Besides, shoot Mn concentrations of bread wheat cultivars were approximately 30% higher than the durum wheat varieties (Table 3.37).

According to the ANOVA results, main effects of B toxicity, cultivar and the interaction of these two stress factors significantly affected the shoot Cu concentration (Table 3.35). Although the application of moderate B toxicity did not affect shoot Cu concentration, severe B toxicity treatment significantly increased it. Shoot Cu concentration results were not consistent between bread and durum wheat (Table 3.37).

Table 3.37: Selected micronutrient concentrations of 40-days-old bread wheat (*Triticum aestivum* cv. Nusrat and Adana-99) and durum wheat (*Triticum durum* cv. Saricanak and Fuatbey-2000) grown in soil culture at various levels of soil salinity stress (control: 3.5 $dS.m^{-1}$ EC_e; medium: 6.0 $dS.m^{-1}$ EC_e; high: 8.5 $dS.m^{-1}$ EC_e) and B supply (adequate: 0 $mg.kg^{-1}$ H_3BO_3; moderate: 15 $mg.kg^{-1}$ H_3BO_3; severe: 30 $mg.kg^{-1}$ H_3BO_3). Data points represents the means and standard deviations of 4 replicates.

B Supply	Soil Salinity	Cultivar	Fe Conc. ($mg.kg^{-1}$)	Zn Conc. ($mg.kg^{-1}$)	Mn Conc. ($mg.kg^{-1}$)	Cu Conc. ($mg.kg^{-1}$)
Adequate B	Control	Nusrat	128 ± 9	29 ± 5	70 ± 6	11.4 ± 0.9
		Adana-99	126 ± 8	25 ± 2	70 ± 6	13.4 ± 0.8
		Saricanak	108 ± 4	23 ± 2	60 ± 10	12.5 ± 0.7
		Fuatbey-00	109 ± 16	25 ± 4	52 ± 8	12.8 ± 0.8
	Medium	Nusrat	82 ± 10	32 ± 5	99 ± 16	12.1 ± 0.4
		Adana-99	78 ± 6	27 ± 3	84 ± 14	12.7 ± 0.2
		Saricanak	56 ± 7	25 ± 6	69 ± 6	12.0 ± 1.1
		Fuatbey-00	66 ± 3	28 ± 4	68 ± 10	13.3 ± 1.4
	High	Nusrat	77 ± 14	25 ± 5	94 ± 19	10.8 ± 0.7
		Adana-99	77 ± 15	21 ± 4	92 ± 14	13.8 ± 0.4
		Saricanak	68 ± 9	27 ± 5	77 ± 20	11.3 ± 1.0
		Fuatbey-00	61 ± 3	26 ± 3	66 ± 6	12.6 ± 0.8
Moderate B Toxicity	Control	Nusrat	195 ± 93	27 ± 2	65 ± 2	11.5 ± 0.4
		Adana-99	121 ± 20	21 ± 7	62 ± 7	11.5 ± 1.4
		Saricanak	79 ± 11	18 ± 3	53 ± 5	12.1 ± 2.0
		Fuatbey-00	81 ± 6	18 ± 2	55 ± 17	14.5 ± 2.0
	Medium	Nusrat	107 ± 15	21 ± 2	95 ± 14	12.0 ± 0.5
		Adana-99	121 ± 23	19 ± 2	96 ± 9	13.7 ± 1.1
		Saricanak	86 ± 17	22 ± 4	74 ± 17	12.6 ± 1.2
		Fuatbey-00	93 ± 4	25 ± 2	68 ± 10	14.7 ± 1.2
	High	Nusrat	84 ± 24	24 ± 2	92 ± 6	11.6 ± 1.5
		Adana-99	78 ± 7	18 ± 1	86 ± 10	12.3 ± 0.9
		Saricanak	81 ± 18	20 ± 4	75 ± 15	12.4 ± 1.1
		Fuatbey-00	73 ± 10	21 ± 3	71 ± 12	14.2 ± 0.2
Severe B Toxicity	Control	Nusrat	118 ± 20	26 ± 5	73 ± 5	13.1 ± 0.8
		Adana-99	94 ± 11	21 ± 4	71 ± 4	15.1 ± 0.7
		Saricanak	99 ± 7	22 ± 2	47 ± 2	13.1 ± 0.6
		Fuatbey-00	85 ± 9	20 ± 1	44 ± 7	15.6 ± 0.8
	Medium	Nusrat	84 ± 6	26 ± 2	99 ± 6	12.8 ± 1.0
		Adana-99	85 ± 16	22 ± 1	87 ± 8	14.8 ± 1.0
		Saricanak	79 ± 16	25 ± 4	85 ± 24	14.6 ± 1.8
		Fuatbey-00	68 ± 8	26 ± 3	66 ± 16	14.2 ± 1.6
	High	Nusrat	84 ± 25	30 ± 3	105 ± 12	12.4 ± 1.3
		Adana-99	81 ± 3	21 ± 2	103 ± 5	14.9 ± 0.3
		Saricanak	68 ± 15	21 ± 3	76 ± 12	12.9 ± 0.6
		Fuatbey-00	78 ± 10	22 ± 1	69 ± 5	14.8 ± 1.4

Shoot Fe Conc. $HSD_{0.05}$ (SS; BS; CV; SSxBS; SSxCV; BSxCV; SSxBSxCV)= (10; 10; 12; 23; 28; *n.s.*; 57)
Shoot Zn Conc. $HSD_{0.05}$ (SS; BS; CV; SSxBS; SSxCV; BSxCV; SSxBSxCV)= (1.6; 1.6; 2.1; *n.s.*; 3.7; *n.s.*; 7.3.)
Shoot Mn Conc. $HSD_{0.05}$ (SS; BS; CV; SSxBS; SSxCV; BSxCV; SSxBSxCV)= (6; *n.s.*; 7; *n.s.*; *n.s.*; *n.s.*; *n.s.*)
Shoot Cu Conc. $HSD_{0.05}$ (SS; BS; CV; SSxBS; SSxCV; BSxCV; SSxBSxCV)= (*n.s.*; 0.5; 0.6; *n.s.*; *n.s.*; 1.5; *n.s.*)

According to ANOVA results, the main effects of soil salinity, B toxicity, and cultivar significantly affected concentrations of MDA and total protein (Table 3.38).

Table 3.38: Analysis of variance of the main effects of salinity, B toxicity, and cultivar and their interactions on concentrations of malondialdehyde (MDA), glycine betaine (GB), proline and, protein of 40-days-old cultivars of bread wheat (*Triticum aestivum* cv. Nusrat and Adana-99) and durum wheat (*Triticum durum* cv. Saricanak and Fuatbey-2000) grown in soil media under greenhouse conditions.

Source of Variation	MDA Conc.	GB Conc.	Proline Conc.	Protein Conc.
Soil Salinity (SS)	**	**	**	***
B Supply (BS)	***	*n.s.*	***	***
Cultivar (CV)	**	*n.s.*	*n.s.*	***
SS*BS	*n.s.*	*n.s.*	*n.s.*	*n.s.*
BS*CV	*n.s.*	*n.s.*	*n.s.*	*
SS*CV	*n.s.*	*n.s.*	*n.s.*	*n.s.*
SS*BS*CV	*n.s.*	*n.s.*	*n.s.*	*n.s.*

n.s. Not significant; * $0.01 \leq$ F Pr. < 0.05; ** $0.001 \leq$ F Pr. < 0.01; *** F Pr. < 0.001

Medium and high soil salinity treatments increased MDA concentration by about 20% (Figure 3.24). There is no difference was observed between salinity doses on the effects of MDA concentration (Figure 3.25; Table 3.39)

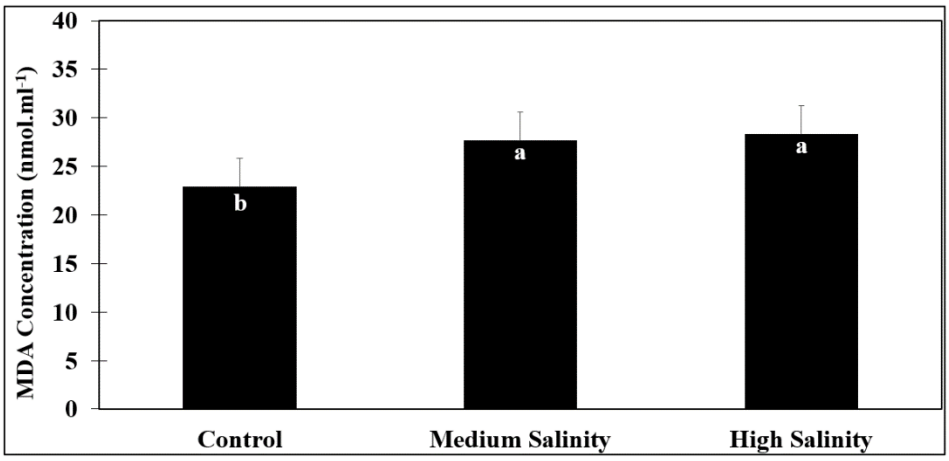

Figure 3.24: Malondialdehyde (MDA) concentrations of 40-days-old wheat grown in soil culture at various levels of soil salinity stress (control: 3.5 $dS.m^{-1}$ EC_e; medium: 6.0 $dS.m^{-1}$ EC_e; high: 8.5 $dS.m^{-1}$ EC_e) under greenhouse conditions. Different letters indicate significant differences between means according to one-way ANOVA and Tukey's HSD test ($p \leq 0.05$).

Severe B toxicity treatment increased MDA concentration by 38%. However, MDA concentration was increased by moderate B toxicity application by 70% (Figure 3.25; Table 3.39).

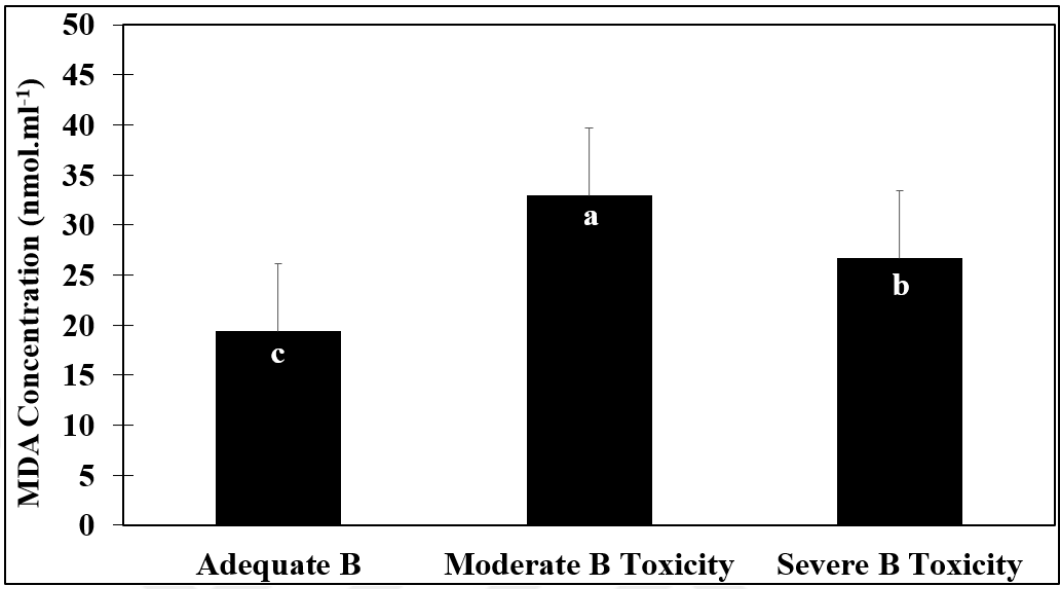

Figure 3.25: Effects of different doses of B supply (adequate: 0 mg.kg^{-1} H_3BO_3; moderate: 15 mg.kg^{-1} H_3BO_3; severe: 30 mg.kg^{-1} H_3BO_3) on malondialdehyde (MDA) concentrations of 40-days-old wheat grown in soil under greenhouse conditions. Different letters indicate significant differences between means according to one-way ANOVA and Tukey's HSD test ($p \leq 0.05$).

Among the cultivars, while the highest concentration of MDA was detected in Nusrat, but the lowest was found in Adana-99 (Figure 3.26; Table 3.39)

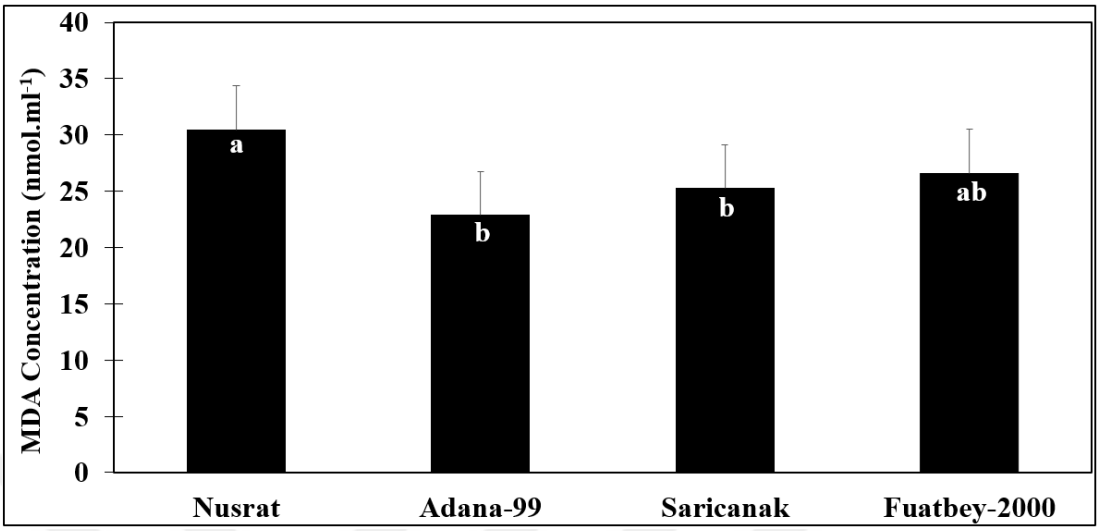

Figure 3.26: Malondialdehyde (MDA) concentrations of 40-days-old bread wheat (*Triticum aestivum* cv. Nusrat and Adana-99) and durum wheat (*Triticum durum* cv. Saricanak and Fuatbey-2000) grown in soil under greenhouse conditions. Different letters indicate significant differences between means according to one-way ANOVA and Tukey's HSD test ($p \leq 0.05$).

Glycine betaine concentration significantly increased in the presence of soil salinity stress (Table 3.38; Figure 3.27). However, there was no difference observed between medium and high salinity applications. Also, the main effects of B toxicity and cultivar did not influence GB concentration (Table 3.39).

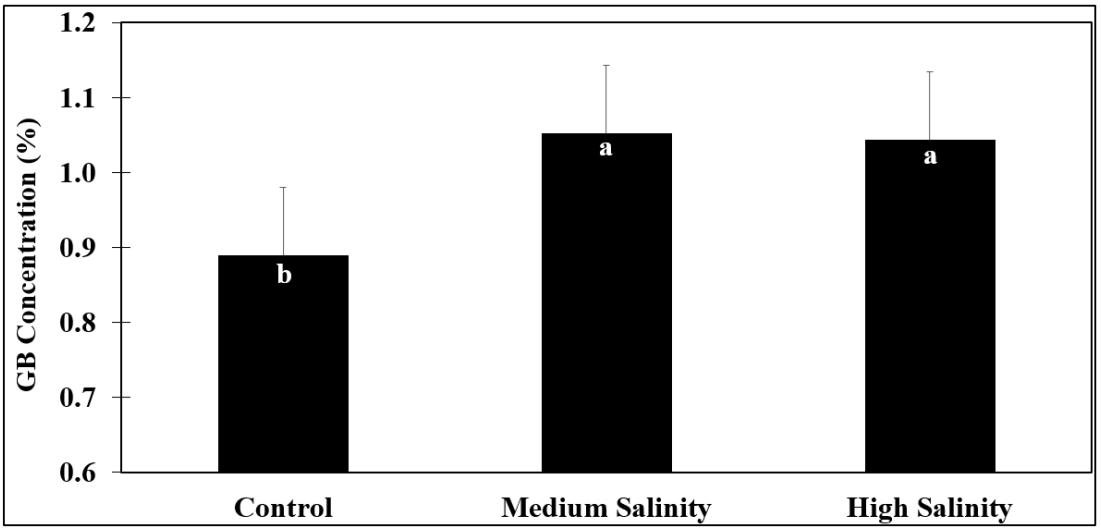

Figure 3.27: Glycine betaine (GB) concentrations of 40-days-old wheat grown in soil at different levels of soil salinity stress (control:e 3.5 dS.m-1 ECe; medium: 6.0 dS.m-1 ECe; high: 8.5 dS.m-1 ECe) under greenhouse conditions. Different letters indicate significant differences between means according to one-way ANOVA and Tukey's HSD test ($p \leq 0.05$).

Moderate and severe B toxicity treatments increased the proline concentration by 63% (Table 3.39). Medium soil salinity treatment did not affect proline concentration, while high salinity application increased proline concentration by 30%.

The main effects of salinity and B toxicity increased protein concentrations at similar levels (Table 3.39). Besides, the highest protein concentration was observed in the durum wheat cultivar Fuatbey-2000. Also, protein concentration in other cultivars was detected as similar between them.

Table 3.39: Effects of various treatments of soil salinity stress (control: 3.5 $dS.m^{-1}$ EC_e; medium: 6.0 $dS.m^{-1}$ EC_e; high: 8.5 $dS.m^{-1}$ EC_e) and B supply (adequate: 0 $mg.kg^{-1}$ H_3BO_3; moderate: 15 $mg.kg^{-1}$ H_3BO_3; severe: 30 $mg.kg^{-1}$ H_3BO_3) on concentrations of malondialdehyde (MDA), glycine betaine (GB), proline, and protein of 40-days-old cultivars of bread wheat (*Triticum aestivum* cv. Nusrat and Adana-99) and durum wheat (*Triticum durum* cv. Saricanak and Fuatbey-2000) grown in soil under greenhouse conditions. Data points represents the means and standard deviations of four replicates.

B Supply	Soil Salinity	Cultivar	MDA Conc. ($nmol.ml^{-1}$)	GB Conc. (%)	Proline Conc. (%)	Protein Conc. (%)
Adequate B	Control	Nusrat	17 ± 6	0.8 ± 0.1	42 ± 3	1.03 ± 0.04
		Adana-99	14 ± 4	0.8 ± 0.2	35 ± 16	1.24 ± 0.09
		Saricanak	17 ± 3	1.0 ± 0.2	30 ± 13	1.11 ± 0.04
		Fuatbey-00	11 ± 3	0.9 ± 0.1	32 ± 14	1.19 ± 0.04
	Medium	Nusrat	20 ± 1	0.9 ± 0.1	35 ± 13	1.22 ± 0.04
		Adana-99	17 ± 5	0.8 ± 0.1	35 ± 25	1.18 ± 0.10
		Saricanak	19 ± 3	1.0 ± 0.2	49 ± 26	1.12 ± 0.14
		Fuatbey-00	26 ± 4	1.2 ± 0.2	32 ± 12	1.29 ± 0.04
	High	Nusrat	26 ± 7	1.2 ± 0.2	27 ± 10	1.22 ± 0.05
		Adana-99	18 ± 5	1.1 ± 0.1	37 ± 20	1.17 ± 0.09
		Saricanak	25 ± 4	0.9 ± 0.1	54 ± 12	1.15 ± 0.01
		Fuatbey-00	22 ± 13	1.2 ± 0.3	46 ± 8	1.30 ± 0.06
Moderate B Toxicity	Control	Nusrat	30 ± 5	0.9 ± 0.1	46 ± 10	1.27 ± 0.18
		Adana-99	23 ± 6	0.7 ± 0.1	49 ± 12	1.11 ± 0.10
		Saricanak	25 ± 8	0.9 ± 0.1	51 ± 16	1.21 ± 0.04
		Fuatbey-00	39 ± 4	0.9 ± 0.1	74 ± 37	1.31 ± 0.03
	Medium	Nusrat	42 ± 17	0.8 ± 0.1	42 ± 4	1.32 ± 0.03
		Adana-99	30 ± 11	0.9 ± 0.1	48 ± 24	1.25 ± 0.04
		Saricanak	33 ± 15	1.1 ± 0.1	58 ± 28	1.26 ± 0.09
		Fuatbey-00	31 ± 7	1.3 ± 0.1	51 ± 19	1.28 ± 0.06
	High	Nusrat	43 ± 16	1.1 ± 0.2	89 ± 48	1.35 ± 0.10
		Adana-99	29 ± 8	1.1 ± 0.3	88 ± 44	1.30 ± 0.02
		Saricanak	35 ± 11	1.1 ± 0.1	71 ± 29	1.26 ± 0.11
		Fuatbey-00	34 ± 3	1.1 ± 0.3	81 ± 28	1.37 ± 0.03
Severe B Toxicity	Control	Nusrat	33 ± 11	0.9 ± 0.3	63 ± 19	1.23 ± 0.17
		Adana-99	20 ± 7	0.6 ± 0.1	47 ± 9	1.14 ± 0.08
		Saricanak	20 ± 5	1.1 ± 0.3	57 ± 20	1.19 ± 0.07
		Fuatbey-00	25 ± 8	1.2 ± 0.4	57 ± 21	1.21 ± 0.04
	Medium	Nusrat	37 ± 2	1.1 ± 0.3	58 ± 19	1.23 ± 0.04
		Adana-99	25 ± 7	1.2 ± 0.3	46 ± 8	1.22 ± 0.15
		Saricanak	23 ± 3	1.3 ± 0.3	71 ± 21	1.32 ± 0.07
		Fuatbey-00	29 ± 6	1.0 ± 0.3	69 ± 21	1.34 ± 0.12
	High	Nusrat	24 ± 5	0.8 ± 0.4	71 ± 36	1.32 ± 0.03
		Adana-99	30 ± 6	1.1 ± 0.7	63 ± 3	1.27 ± 0.08
		Saricanak	30 ± 9	1.2 ± 0.6	65 ± 14	1.27 ± 0.06
		Fuatbey-00	23 ± 8	0.7 ± 0.1	73 ± 24	1.35 ± 0.02

MDA Concentration $HSD_{0.05}$ (SS; BS; CV; SSxBS; SSxCV; BSxCV; SSxBSxCV)= (4; 4; 5; *n.s.*; *n.s.*; *n.s.*; *n.s.*)
GB Concentration $HSD_{0.05}$ (SS; BS; CV; SSxBS; SSxCV; BSxCV; SSxBSxCV)= (0.1; *n.s.*; *n.s.*; *n.s.*; *n.s.*; *n.s.*; *n.s.*)
Proline Concentration $HSD_{0.05}$ (SS; BS; CV; SSxBS; SSxCV; BSxCV; SSxBSxCV)= (10; 10; *n.s.*; *n.s.*; *n.s.*; *n.s.*; *n.s.*)
Protein Concentration $HSD_{0.05}$ (SS; BS; CV; SSxBS; SSxCV; BSxCV; SSxBSxCV)= (0.04; 0.04; 0.05; *n.s.*; *n.s.*; 0.1; *n.s.*)

Table 3.40: Analysis of variance of the main effects of salinity, B toxicity, and cultivar and their interaction on total (A) and specific (B) enzyme activities of superoxide dismutase (SOD), glutathione reductase (GR), ascorbate peroxidase (AP), and catalase (CAT) of 40-days-old cultivars of bread wheat (*Triticum aestivum* cv. Nusrat and Adana-99) and durum wheat (*Triticum durum* cv. Saricanak and Fuatbey-2000) grown in soil media under greenhouse conditions.

	Source of Variation	**SOD**	**GR**	**APX**	**CAT**
(A) Normal Enzyme Activities	Soil Salinity (SS)	*n.s.*	*n.s.*	**	**
	B Supply (BS)	***	***	**	**
	Cultivar (CV)	***	***	**	***
	SS*BS	*n.s.*	*n.s.*	*n.s.*	*n.s.*
	BS*CV	*n.s.*	*n.s.*	*	*
	SS*CV	*n.s.*	*n.s.*	*n.s.*	*n.s.*
	SS*BS*CV	*n.s.*	*n.s.*	*n.s.*	*n.s.*
	Source of Variation	**SOD**	**GR**	**APX**	**CAT**
(B) Specific Enzyme Activities	Soil Salinity (SS)	**	*n.s.*	***	*n.s.*
	B Supply (BS)	**	***	*n.s.*	**
	Cultivar (CV)	*n.s.*	*n.s.*	**	***
	SS*BS	*n.s.*	*	*n.s.*	*n.s.*
	BS*CV	*	*n.s.*	*	*n.s.*
	SS*CV	*n.s.*	*n.s.*	*n.s.*	*n.s.*
	SS*BS*CV	*	*n.s.*	*n.s.*	*n.s.*

n.s. Not significant; * 0.01 ≤ F Pr. < 0.05; ** 0.001 ≤ F Pr. < 0.01; *** F Pr. < 0.001

Severe B toxicity application to wheat increased the SOD enzyme total activity (Table 3.40; Table 3.41). Fuatbey-2000 SOD enzyme activity is 7% higher than other wheat cultivars. While GR enzyme activity decreased by 10% with the treatment of moderate B toxicity, it decreased by 18% with the application of severe B toxicity (Table 3.40; Table 3.41). Fuatbey-2000 GR enzyme activity was found to be significantly higher than other wheat cultivars. Medium salinity stress tended to decrease APX activity (Table 3.40; Table 3.41). However, high salinity stress significantly reduced APX activity. Moderate and severe B toxicity treatments significantly reduced APX enzyme activity. Fuatbey-2000 was found to have the highest APX enzyme activity among other wheat varieties. Medium and high salinity stress tended to increase CAT enzyme activity (Table 3.40; Table 3.41). Moderate B toxicity increased CAT enzyme activity in wheat, but severe B toxicity significantly decreased CAT enzyme activity. Catalase enzyme activity of Fuatbey-2000 was found to be significantly higher than other wheat varieties.

Table 3.41: Total antioxidant enzyme; superoxide dismutase (SOD), glutathione reductase (GR), ascorbate peroxidase (AP), and catalase (CAT) activities of 40-days-old bread wheat (*Triticum aestivum* cv. Nusrat and Adana-99) and durum wheat (*Triticum durum* cv. Saricanak and Fuatbey-2000) grown in soil culture at various levels of soil salinity stress (control: 3.5 $dS.m^{-1}$ EC_e; medium: 6.0 $dS.m^{-1}$ EC_e; high: 8.5 $dS.m^{-1}$ EC_e) and B supply (adequate: 0 $mg.kg^{-1}$ H_3BO_3; moderate: 15 $mg.kg^{-1}$ H_3BO_3; severe: 30 $mg.kg^{-1}$ H_3BO_3). Data points represents the means and standard deviations of four replicates.

B Supply	Soil Salinity	Cultivar	SOD Act. (U.g^{-1} FW)	GR Act. (-µmol NADPH g^{-1} FW min^{-1})	APX Act. (-µmol H_2O_2 g^{-1} FW min^{-1})	CAT Act. (-mmol H_2O_2 g^{-1} FW min^{-1})
Adequate B	Control	Nusrat	119 ± 3	2.4 ± 0.2	86 ± 9	4.1 ± 0.8
		Adana-99	105 ± 5	2.7 ± 0.9	81 ± 10	5.2 ± 0.9
		Saricanak	111 ± 12	2.7 ± 0.5	92 ± 9	5.1 ± 0.3
		Fuatbey-00	119 ± 6	3.3 ± 0.3	126 ± 27	6.2 ± 0.6
	Medium	Nusrat	115 ± 8	2.7 ± 0.4	87 ± 15	5.4 ± 0.5
		Adana-99	111 ± 6	2.9 ± 0.6	92 ± 20	5.4 ± 0.8
		Saricanak	111 ± 5	2.5 ± 0.3	67 ± 15	5.2 ± 0.6
		Fuatbey-00	118 ± 4	3.7 ± 0.6	94 ± 22	6.3 ± 0.7
	High	Nusrat	113 ± 9	3.3 ± 1.1	69 ± 5	4.6 ± 0.5
		Adana-99	114 ± 10	3.0 ± 0.3	82 ± 14	5.1 ± 0.4
		Saricanak	115 ± 4	3.2 ± 0.9	82 ± 5	5.3 ± 0.5
		Fuatbey-00	115 ± 5	3.4 ± 0.5	93 ± 8	6.5 ± 0.6
Moderate B Toxicity	Control	Nusrat	109 ± 9	2.6 ± 0.4	105 ± 6	5.3 ± 1.1
		Adana-99	112 ± 4	2.6 ± 0.3	92 ± 11	4.6 ± 0.3
		Saricanak	116 ± 17	2.4 ± 0.7	97 ± 12	5.1 ± 0.7
		Fuatbey-00	114 ± 3	2.9 ± 0.2	111 ± 8	6.3 ± 1.1
	Medium	Nusrat	106 ± 9	2.9 ± 0.4	86 ± 2	5.3 ± 0.8
		Adana-99	112 ± 7	2.8 ± 0.3	92 ± 12	5.6 ± 0.6
		Saricanak	116 ± 2	2.7 ± 0.2	97 ± 14	5.6 ± 0.5
		Fuatbey-00	118 ± 3	3.2 ± 0.4	106 ± 6	6.3 ± 0.6
	High	Nusrat	119 ± 8	2.7 ± 0.2	88 ± 5	5.6 ± 1.1
		Adana-99	113 ± 10	2.9 ± 0.2	109 ± 10	5.5 ± 0.4
		Saricanak	119 ± 6	2.5 ± 0.2	82 ± 23	5.6 ± 0.7
		Fuatbey-00	126 ± 4	2.8 ± 0.1	101 ± 40	6.7 ± 0.9
Severe B Toxicity	Control	Nusrat	118 ± 8	2.3 ± 0.3	97 ± 11	4.4 ± 0.5
		Adana-99	121 ± 5	2.5 ± 0.6	113 ± 25	4.7 ± 0.6
		Saricanak	118 ± 6	2.5 ± 0.3	102 ± 7	5.4 ± 0.9
		Fuatbey-00	126 ± 6	2.7 ± 0.3	100 ± 14	5.2 ± 0.4
	Medium	Nusrat	121 ± 5	2.6 ± 0.2	90 ± 14	4.9 ± 0.3
		Adana-99	115 ± 8	2.3 ± 0.4	103 ± 20	5.1 ± 0.8
		Saricanak	121 ± 8	2.5 ± 0.2	104 ± 11	5.9 ± 0.8
		Fuatbey-00	134 ± 17	2.6 ± 0.2	104 ± 8	5.7 ± 0.5
	High	Nusrat	118 ± 3	2.5 ± 0.4	90 ± 14	5.1 ± 0.5
		Adana-99	117 ± 7	2.2 ± 0.2	109 ± 17	4.7 ± 0.4
		Saricanak	115 ± 1	2.2 ± 0.2	89 ± 6	5.2 ± 0.4
		Fuatbey-00	131 ± 9	2.5 ± 0.1	89 ± 7	5.7 ± 0.3

SOD Act. $HSD_{0.05}$ (SS; BS; CV; SSxBS; SSxCV; BSxCV; SSxBSxCV)= (*n.s.*; 4; 5; *n.s.*; *n.s.*; *n.s.*; *n.s.*)
GR Act.$HSD_{0.05}$ (SS; BS; CV; SSxBS; SSxCV; BSxCV; SSxBSxCV)= (*n.s.*; 0.2; 0.2; *n.s.*; *n.s.*; *n.s.*; *n.s.*)
APX Act. $HSD_{0.05}$ (SS; BS; CV; SSxBS; SSxCV; BSxCV; SSxBSxCV)= (8; 8; 10; *n.s.*; *n.s.*; 20; *n.s.*)
CAT Act. $HSD_{0.05}$ (SS; BS; CV; SSxBS; SSxCV; BSxCV; SSxBSxCV)= (0.3; 0.3; 0.4; *n.s.*; *n.s.*; 0.9; *n.s.*)

According to the ANOVA results, soil salinity, B toxicity, B toxicity*cultivar, and triple interaction significantly affected the specific (sp.) SOD enzyme activity (Table 3.40). Specific SOD enzyme activity decreased significantly in the presence of salinity stress. Also, sp. SOD enzyme activity significantly reduced in wheat plants treated with moderate B toxicity. However, the application of severe B toxicity did not affect sp. SOD enzyme activity (Table 3.40; Table 3.42).

Moderate and severe B toxicity treatment gradually decreased the sp. GR enzyme activity (Table 3.40; Table 3.42). Medium and high salinity treatment reduced the sp. APX enzyme activity by 18% approximately (Table 3.40; Table 3.42).

Moderate B toxicity treatment did not affect sp. CAT enzyme activity, but severe B toxicity treatment significantly reduced it (Table 3.40; Table 3.42). Fuatbey-2000 sp. CAT enzyme activity was found to be higher than other wheat cultivars. Besides, the lowest sp. CAT enzyme activity was detected in Nusrat.

Table 3.42: Effects of different levels of salinity (control: 3.5 $dS.m^{-1}$ EC_e; medium: 6.0 $dS.m^{-1}$ EC_e; high: 8.5 $dS.m^{-1}$ EC_e) and B supply (adequate: 0 $mg.kg^{-1}$ H_3BO_3; moderate: 15 $mg.kg^{-1}$ H_3BO_3; severe: 30 $mg.kg^{-1}$ H_3BO_3) on specific antioxidant enzyme; superoxide dismutase (SOD), glutathione reductase (GR), ascorbate peroxidase (AP), and catalase (CAT) activities of 40-days-old cultivars of bread wheat (*Triticum aestivum* cv. Nusrat and Adana-99) and durum wheat (*Triticum durum* cv. Saricanak and Fuatbey-2000) grown in soil under greenhouse conditions. Data points represents the means and standard deviations of four replicates.

B Supply	Soil Salinity	Cultivar	Sp. SOD Act. (U.mg^{-1} prt.)	Sp. GR Act. (-nmol [NADPH] mg^{-1} prt.min^{-1})	Sp. APX Act. (-µmol H_2O_2 g^{-1} FW min^{-1})	Sp. CAT Act. (-mmol H_2O_2 g^{-1} FW min^{-1})
Adequate B	Control	Nusrat	11.6 ± 0.4	229 ± 8	8.4 ± 1.3	0.39 ± 0.07
		Adana-99	8.6 ± 0.8	218 ± 59	6.8 ± 1.2	0.42 ± 0.06
		Saricanak	10.0 ± 1.3	242 ± 42	8.3 ± 0.8	0.47 ± 0.04
		Fuatbey-00	10.0 ± 0.7	280 ± 28	10.7 ± 2.4	0.52 ± 0.05
	Medium	Nusrat	9.4 ± 0.9	221 ± 35	7.2 ± 1.5	0.44 ± 0.05
		Adana-99	9.4 ± 1.2	241 ± 41	7.8 ± 1.4	0.45 ± 0.04
		Saricanak	10.0 ± 1.4	223 ± 36	5.9 ± 0.7	0.46 ± 0.04
		Fuatbey-00	9.2 ± 0.1	283 ± 40	7.3 ± 2.0	0.49 ± 0.06
	High	Nusrat	9.3 ± 0.9	273 ± 85	5.6 ± 0.4	0.38 ± 0.04
		Adana-99	9.8 ± 1.1	257 ± 19	7.0 ± 0.8	0.43 ± 0.02
		Saricanak	10.0 ± 0.3	280 ± 72	7.1 ± 0.4	0.46 ± 0.05
		Fuatbey-00	8.9 ± 0.3	261 ± 35	7.2 ± 0.7	0.51 ± 0.06
Moderate B Toxicity	Control	Nusrat	8.7 ± 1.3	205 ± 45	8.4 ± 1.0	0.42 ± 0.04
		Adana-99	10.1 ± 1.1	234 ± 13	8.3 ± 0.6	0.41 ± 0.05
		Saricanak	9.5 ± 1.2	194 ± 47	8.0 ± 0.8	0.42 ± 0.05
		Fuatbey-00	8.7 ± 0.4	220 ± 16	8.5 ± 0.8	0.48 ± 0.10
	Medium	Nusrat	8.0 ± 0.9	217 ± 30	6.5 ± 0.3	0.40 ± 0.06
		Adana-99	8.9 ± 0.7	221 ± 27	7.3 ± 0.9	0.45 ± 0.04
		Saricanak	9.3 ± 0.8	212 ± 25	7.7 ± 0.7	0.44 ± 0.01
		Fuatbey-00	9.3 ± 0.6	253 ± 23	8.3 ± 0.7	0.49 ± 0.05
	High	Nusrat	8.9 ± 1.0	201 ± 24	6.5 ± 0.8	0.41 ± 0.05
		Adana-99	8.7 ± 0.9	224 ± 13	8.4 ± 0.8	0.43 ± 0.03
		Saricanak	9.5 ± 0.9	199 ± 19	6.5 ± 1.6	0.45 ± 0.07
		Fuatbey-00	9.2 ± 0.5	204 ± 10	7.3 ± 2.8	0.49 ± 0.07
Severe B Toxicity	Control	Nusrat	9.8 ± 2.1	193 ± 30	8.0 ± 1.5	0.36 ± 0.01
		Adana-99	10.7 ± 0.9	216 ± 46	9.9 ± 1.9	0.41 ± 0.04
		Saricanak	10.0 ± 1.0	208 ± 22	8.6 ± 1.0	0.45 ± 0.06
		Fuatbey-00	10.5 ± 0.4	221 ± 20	8.3 ± 1.2	0.43 ± 0.03
	Medium	Nusrat	9.8 ± 0.6	209 ± 19	7.3 ± 1.2	0.40 ± 0.02
		Adana-99	9.6 ± 1.2	192 ± 22	8.5 ± 0.8	0.42 ± 0.05
		Saricanak	9.2 ± 0.3	190 ± 22	7.8 ± 0.8	0.44 ± 0.06
		Fuatbey-00	10.1 ± 2.2	197 ± 10	7.8 ± 0.3	0.43 ± 0.02
	High	Nusrat	8.9 ± 0.3	188 ± 37	6.8 ± 1.1	0.39 ± 0.04
		Adana-99	9.2 ± 0.5	175 ± 13	8.6 ± 1.2	0.37 ± 0.01
		Saricanak	9.1 ± 0.4	173 ± 10	7.0 ± 0.3	0.41 ± 0.01
		Fuatbey-00	9.7 ± 0.7	185 ± 11	6.6 ± 0.6	0.42 ± 0.03

SOD Act. $HSD_{0.05}$ (SS; BS; CV; SSxBS; SSxCV; BSxCV; SSxBSxCV)= (0.5; 0.5; *n.s.*; *n.s.*; *n.s.*; 1.5; 3.0)
GR Act.$HSD_{0.05}$ (SS; BS; CV; SSxBS; SSxCV; BSxCV; SSxBSxCV)= (*n.s.*; 17; *n.s.*; 40; *n.s.*; *n.s.*; *n.s.*)
APX Act. $HSD_{0.05}$ (SS; BS; CV; SSxBS; SSxCV; BSxCV; SSxBSxCV)= (0.6; *n.s.*; 0.7; *n.s.*; *n.s.*; 2.0; *n.s.*)
CAT Act. $HSD_{0.05}$ (SS; BS; CV; SSxBS; SSxCV; BSxCV; SSxBSxCV)= (*n.s.*; 0.02; 0.02; *n.s.*; *n.s.*; *n.s.*; *n.s.*)

3.2.3. Third Soil Experiment Results

The third soil experiment was conducted to study the effects of the interaction of different levels of salinity stress with various doses of B toxicity on shoot growth, yield parameters, shoot and grain accumulation of selected minerals, K/Na ratio, shoot symptom score, and relative electrolyte leakage in two bread wheat and two durum wheat cultivars in soil culture under greenhouse conditions. The difference of this soil experiment from the previous soil experiment is that the salinity stress and B toxicity doses are slightly reduced, and the wheat crops were grown to the end of maturity.

Under the non-saline and saline conditions, moderate and severe B toxicity was caused chlorosis and necrosis in mature leaves of experimental plants beginning from leaf tips and margins and impaired growth and development (Figure 3.28). Also, especially in Saricanak durum wheat cultivar, increasing salinity was reduced plant growth and tillering under all three B toxicity conditions.

The cultivar of bread wheat Adana-99 and durum wheat Saricanak were recognized as in the anthesis stage of wheat development in the figure (Figure 3.28). But Nusrat development differed dependent on the salinity and B toxicity intensity. According to this situation, Nusrat growth varied between booting and heading status. Unlike other cultivars, the development of Fuatbey-2000 was very late, and plants were seen at the flag leaf stage.

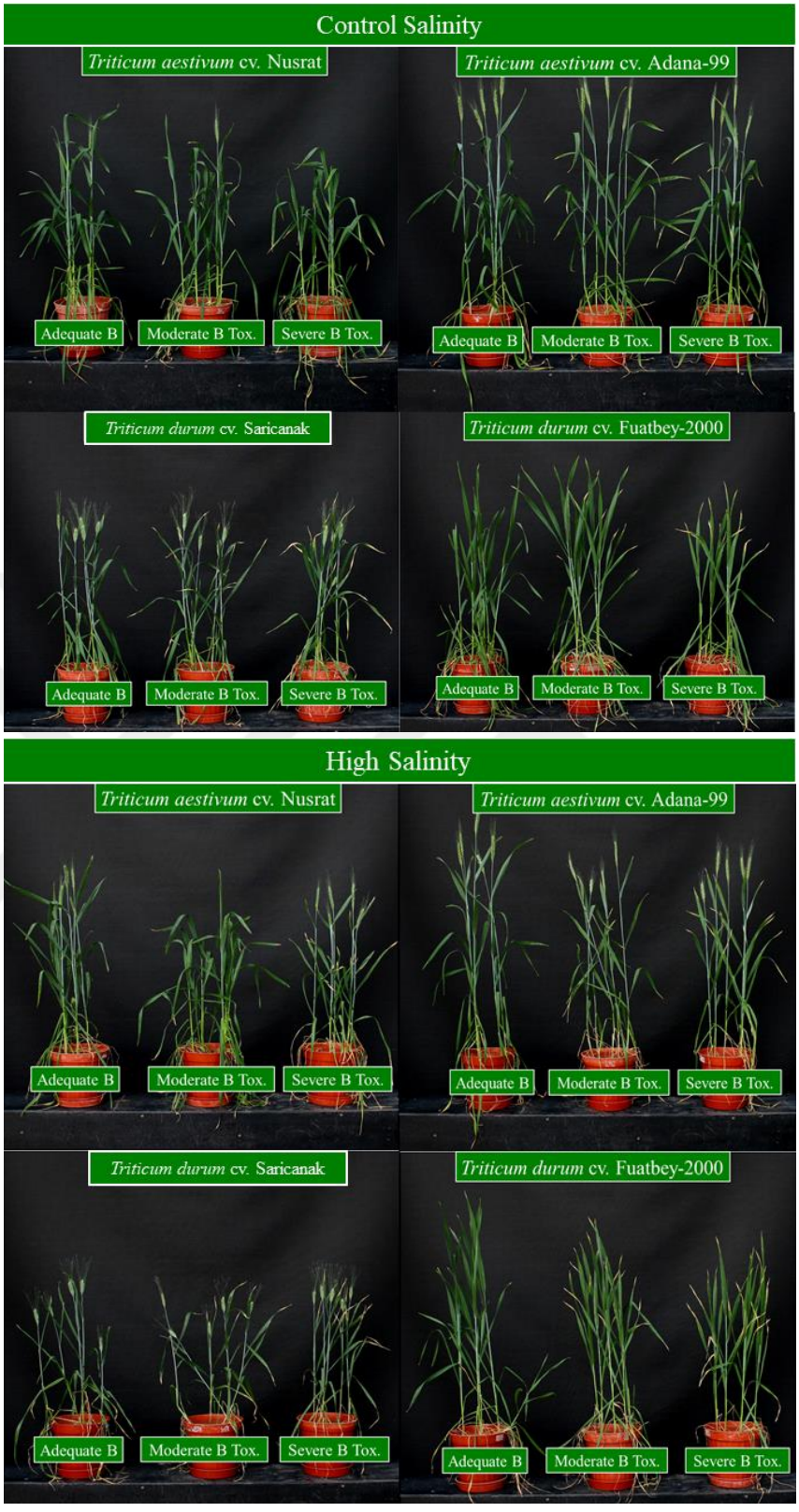

Figure 3.28: Shoot growth, tillering, and booting of 90-days-old cultivars of bread wheat (*Triticum aestivum* cv. Nusrat and Adana-99) and durum wheat (*Triticum durum* cv. Saricanak and Fuatbey-2000) grown in soil at three different levels of soil salinity stress (control: 3.5 $dS.m^{-1}$ EC_e; medium: 5.5 $dS.m^{-1}$ EC_e; high: 7.5 $dS.m^{-1}$ EC_e) and B supply (adequate: 0 $mg.kg^{-1}$ H_3BO_3; moderate: 10 $mg.kg^{-1}$ H_3BO_3; severe: 20 $mg.kg^{-1}$ H_3BO_3).

Boron toxicity treatment significantly increased the symptom score of wheat plants (Table 3.43). It was determined that Saricanak, which is a variety of durum wheat, had the highest symptom score. Nusrat, a bread wheat variety, had the lowest symptom score. There was no significant difference in symptom score between Adana-99 and Fuatbey-2000 varieties.

Table 3.43: Symptom score of 89-days-old bread wheat (*Triticum aestivum* cv. Nusrat and Adana-99) and durum wheat (*Triticum durum* cv. Saricanak and Fuatbey-2000) grown in soil culture at different levels of soil salinity stress (control: 3.5 $dS.m^{-1}$ EC_e; medium: 5.5 $dS.m^{-1}$ EC_e; high: 7.5 $dS.m^{-1}$ EC_e) and B supply (adequate: 0 $mg.kg^{-1}$ H_3BO_3; moderate: 10 $mg.kg^{-1}$ H_3BO_3; severe: 20 $mg.kg^{-1}$ H_3BO_3). Symptom score shows visual chlorosis and necrosis starting on leaf tips and margins of second youngest leaves.

Soil Salinity	**Cultivar**	**Symptom Score***		
		Adequate B	Moderate B Tox.	Severe B Tox.
Control	Nusrat	0.3 ± 0.5	3.3 ± 1.0	5.0 ± 1.2
	Adana-99	0.8 ± 0.5	4.3 ± 0.5	5.5 ± 0.6
	Saricanak	1.3 ± 0.5	5.5 ± 0.6	7.3 ± 1.0
	Fuatbey-2000	1.8 ± 1.7	4.8 ± 1.0	5.5 ± 1.0
Medium	Nusrat	1.5 ± 0.6	3.3 ± 1.0	4.5 ± 0.6
	Adana-99	1.3 ± 0.5	4.3 ± 0.5	6.0 ± 0.0
	Saricanak	2.0 ± 0.8	5.5 ± 1.0	8.0 ± 0.0
	Fuatbey-2000	2.0 ± 0.0	4.5 ± 1.0	6.5 ± 1.0
High	Nusrat	1.5 ± 0.6	3.3 ± 1.0	5.0 ± 0.8
	Adana-99	1.5 ± 0.6	4.3 ± 0.5	5.8 ± 0.5
	Saricanak	2.0 ± 0.0	4.8 ± 1.0	6.5 ± 1.0
	Fuatbey-2000	2.0 ± 0.0	5.0 ± 1.2	5.5 ± 0.6

Symptom score $HSD_{0.05}$ (SS; BT; CV; SSxBT; SSxCV; BTxCV; SSxBTxCV)= (*n.s.*; 0.4; 0.5; *n.s.*; *n.s.*; 0.8; *n.s.*)
* Depending on severity of leaf tip and margin necrosis, plants for given scores ranging from 0 (non) to 10 (most).

The main effects of salinity stress and B toxicity significantly increased relative electrolyte leakage (REL) (Figure 3.29). It was also detected that the interaction of these stress factors has an additive effect on the REL of wheat cultivars except Saricanak. Relative electrolyte leakage differed in all four wheat varieties. The lowest REL was determined in Fuatbey-2000 durum wheat cultivar. The REL amount of Nusrat and Adana-99, which are bread wheat varieties, were found to be 107% and 200% higher than Fuatbey-2000, respectively. Besides, it was determined that the REL amount of the durum wheat cultivar Saricanak was 425% higher than Fuatbey-2000.

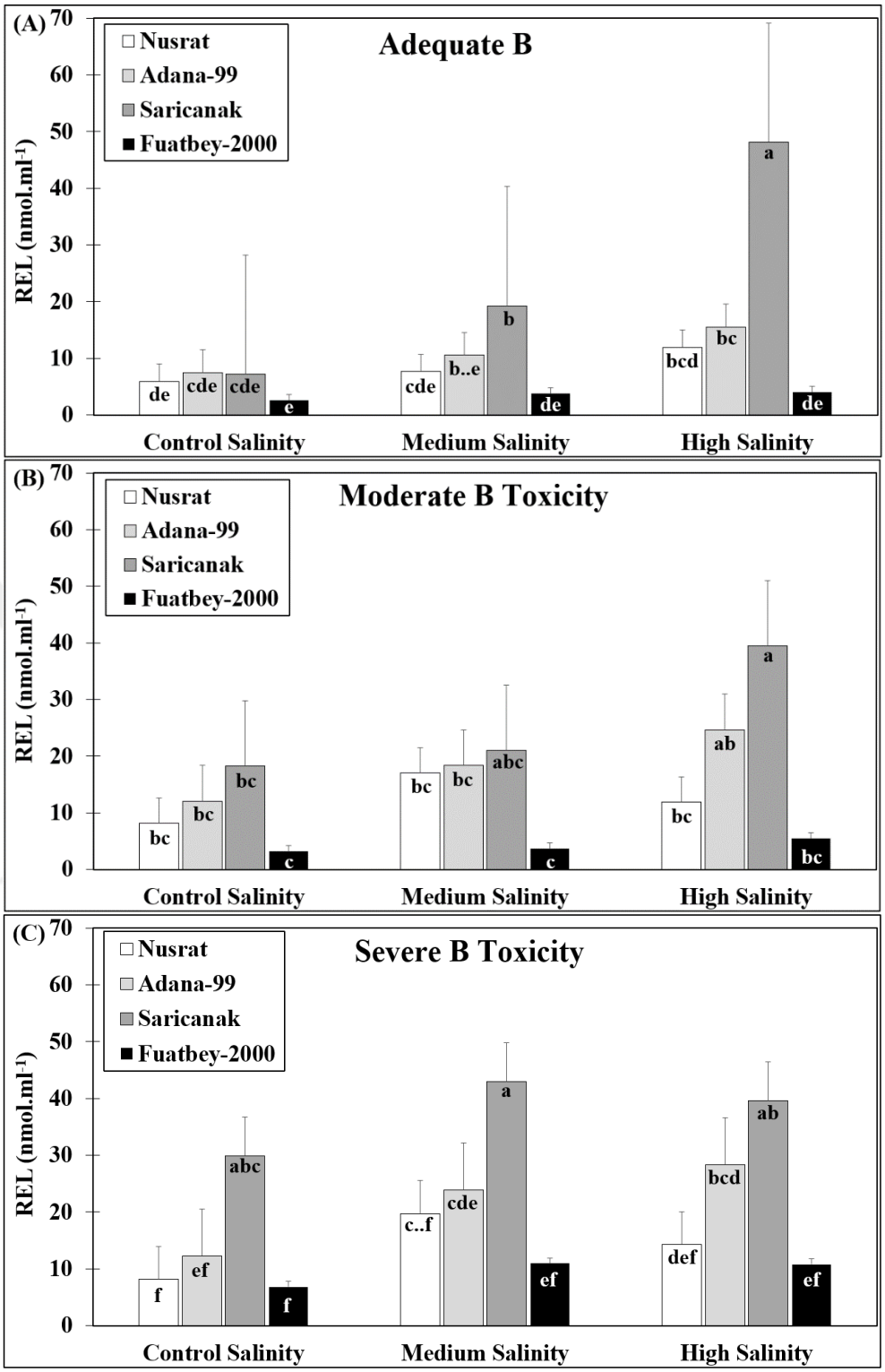

Figure 3.29: Effects of (A) adequate: 0 mg.kg^{-1}, (B) moderate: 10 mg.kg^{-1} and, (C) severe: 20 mg.kg^{-1} H_3BO_3 treatments on relative electrolyte leakage (REL) of 114-days-old bread wheat (*Triticum aestivum* cv. Nusrat and Adana-99) and durum wheat (*Triticum durum* cv. Saricanak and Fuatbey-2000) grown in soil at different levels of soil salinity stress (control: 3.5 dS.m^{-1} EC_e; medium: 5.5 dS.m^{-1} EC_e; high: 7.5 dS.m^{-1} EC_e) under greenhouse conditions. Different letters indicate significant differences between means according to two-way ANOVA and Tukey's HSD test ($p \leq 0.05$).

Table 3.44: Three-way analysis of variance of the effects of salinity, B toxicity, and cultivar and their interaction on straw and grain concentrations of selected nutrients of completely mature cultivars of bread wheat (*Triticum aestivum* cv. Nusrat and Adana-99) and durum wheat (*Triticum durum* cv. Saricanak and Fuatbey-2000) grown in soil under greenhouse conditions: degrees of freedom, F value probabilities, and Tukey's HSD test scores.

Source of Variation	**DF**	**Straw B Conc.**		**Straw Na Conc.**		**Straw K Conc.**	
		F Pr.	$HSD_{0.05}$	F Pr.	$HSD_{0.05}$	F Pr.	$HSD_{0.05}$
Soil Salinity (SS)	2	<0.001	21	<0.001	0.1	<0.001	0.1
B Supply (BS)	2	<0.001	21	0.0101	0.1	<0.001	0.1
Cultivar (CV)	3	<0.001	27	<0.001	0.1	<0.001	0.2
SS*BS	4	0.0094	50	0.3085	0.2	0.9482	0.3
BS*CV	6	<0.001	60	<0.001	0.3	<0.001	0.4
SS*CV	6	<0.001	60	0.3483	0.3	<0.001	0.4
SS*BS*CV	12	0.0006	123	0.8745	0.6	0.0575	0.8
Source of Variation	**DF**	**Grain B Conc.**		**Grain Na Conc.**		**Grain K Conc.**	
		F Pr.	$HSD_{0.05}$	F Pr.	$HSD_{0.05}$	F Pr.	$HSD_{0.05}$
Soil Salinity (SS)	2	0.1587	0.4	<0.001	18	0.0197	0.03
B Supply (BS)	2	<0.001	0.4	0.0439	18	<0.001	0.03
Cultivar (CV)	3	<0.001	0.5	<0.001	48	<0.001	0.04
SS*BS	4	0.35	0.9	0.2212	89	0.0169	0.07
BS*CV	6	0.04	1.2	<0.001	107	<0.001	0.08
SS*CV	6	<0.001	1.2	0.2113	107	<0.001	0.08
SS*BS*CV	12	0.0064	2.6	0.0135	231	0.0086	0.17

Medium salinity stress application reduced straw B concentration by 9%, while high salinity caused a 15% decrease (Table 3.44; Table 3.45). Treatments of moderate and severe B toxicity to wheat were increased significantly the straw B concentration. Considering the results in terms of salinity and B toxicity interaction, it was determined that straw B concentration decreased in the presence of salinity stress in plants treated with B toxicity. Saricanak and Fuatbey-2000 had a significantly higher straw B concentration than the bread wheat cultivars.

Medium and high salinity significantly increased straw Na concentration in all four wheat varieties (Table 3.44; Table 3.45). Moderate and severe B toxicity application did not affect straw Na concentration. The durum wheat varieties Saricanak and Fuatbey-2000 had a significantly higher straw Na concentration than the bread wheat cultivars.

Straw K concentration decreased by about 13% in plants treated with medium and high soil salinity (Table 3.44; Table 3.45). Moderate B toxicity treatment increased straw K concentration by 9%, while severe B toxicity treatment increased 17%. Bread wheat varieties Nusrat and Adana-99 were found to have significantly higher straw K concentrations compared to durum wheat varieties.

Table 3.45: Straw B, Na, K concentrations of completely mature bread wheat (*Triticum aestivum* cv. Nusrat and Adana-99) and durum wheat (*Triticum durum* cv. Saricanak and Fuatbey-2000) grown in soil at different levels of soil salinity stress (control: 3.5 $dS.m^{-1}$ EC_e; medium: 5.5 $dS.m^{-1}$ EC_e; high: 7.5 $dS.m^{-1}$ EC_e) and B supply (adequate: 0 $mg.kg^{-1}$ H_3BO_3; moderate: 10 $mg.kg^{-1}$ H_3BO_3; severe: 20 $mg.kg^{-1}$ H_3BO_3). Data points represents the means and standard deviations of four replicates.

B Supply	Soil Salinity	Cultivar	Straw Nutrient Concentrations		
			B ($mg.kg^{-1}$)	Na (%)	K (%)
Adequate B	Control	Nusrat	12 ± 2	0.1 ± 0.0	3.3 ± 0.6
		Adana-99	11 ± 3	0.1 ± 0.0	3.1 ± 0.2
		Saricanak	17 ± 3	0.5 ± 0.0	2.9 ± 0.3
		Fuatbey-00	15 ± 3	0.5 ± 0.0	2.3 ± 0.0
	Medium	Nusrat	14 ± 4	0.5 ± 0.1	3.5 ± 0.2
		Adana-99	14 ± 2	0.9 ± 0.1	2.5 ± 0.1
		Saricanak	19 ± 4	1.8 ± 0.2	2.2 ± 0.4
		Fuatbey-00	16 ± 1	1.8 ± 0.2	1.9 ± 0.1
	High	Nusrat	15 ± 4	1.2 ± 0.2	3.7 ± 0.2
		Adana-99	15 ± 4	1.3 ± 0.2	2.9 ± 0.2
		Saricanak	22 ± 3	2.8 ± 0.7	1.8 ± 0.5
		Fuatbey-00	13 ± 3	2.3 ± 0.1	1.5 ± 0.1
Moderate B Toxicity	Control	Nusrat	204 ± 6	0.1 ± 0.1	3.5 ± 0.3
		Adana-99	207 ± 13	0.1 ± 0.0	3.3 ± 0.1
		Saricanak	308 ± 14	0.5 ± 0.1	3.4 ± 0.2
		Fuatbey-00	274 ± 28	0.4 ± 0.0	2.3 ± 0.2
	Medium	Nusrat	184 ± 3	0.4 ± 0.1	3.9 ± 0.2
		Adana-99	184 ± 15	0.5 ± 0.1	3.2 ± 0.1
		Saricanak	242 ± 20	1.6 ± 0.2	2.3 ± 0.5
		Fuatbey-00	202 ± 35	1.5 ± 0.2	1.8 ± 0.1
	High	Nusrat	177 ± 25	1.0 ± 0.2	4.0 ± 0.5
		Adana-99	167 ± 4	1.3 ± 0.1	3.3 ± 0.1
		Saricanak	202 ± 18	2.7 ± 0.5	1.7 ± 0.2
		Fuatbey-00	239 ± 21	2.2 ± 0.2	1.7 ± 0.3
Severe B Toxicity	Control	Nusrat	462 ± 55	0.1 ± 0.0	3.8 ± 0.3
		Adana-99	468 ± 49	0.1 ± 0.0	3.6 ± 0.1
		Saricanak	754 ± 105	0.6 ± 0.1	2.8 ± 0.2
		Fuatbey-00	553 ± 55	0.4 ± 0.0	2.9 ± 0.1
	Medium	Nusrat	412 ± 34	0.4 ± 0.0	4.2 ± 0.2
		Adana-99	548 ± 48	0.6 ± 0.1	3.5 ± 0.2
		Saricanak	616 ± 113	1.9 ± 0.3	2.1 ± 0.4
		Fuatbey-00	546 ± 21	1.6 ± 0.2	2.2 ± 0.3
	High	Nusrat	450 ± 40	1.0 ± 0.1	4.1 ± 0.4
		Adana-99	434 ± 117	1.1 ± 0.3	3.8 ± 0.2
		Saricanak	490 ± 88	2.9 ± 0.1	2.0 ± 0.3
		Fuatbey-00	577 ± 61	2.3 ± 0.2	1.8 ± 0.3

Straw B Conc. $HSD_{0.05}$ (SS; BS; CV; SSxBS; SSxCV; BSxCV; SSxBSxCV)= (22; 22; 27; 49; 60; 63; 123)
Straw Na Conc. $HSD_{0.05}$ (SS; BS; CV; SSxBS; SSxCV; BSxCV; SSxBSxCV)= (0.1; 0.1; 0.1; *n.s.*; 0.3; *n.s.*; *n.s.*)
Straw K Conc. $HSD_{0.05}$ (SS; BS; CV; SSxBS; SSxCV; BSxCV; SSxBSxCV)= (0.1; 0.1; 0.2; *n.s.*; 0.4; 0.4.; *n.s.*)

Nusrat and Adana-99, which are bread wheat varieties, had a higher K/Na ratio in all treatments than durum wheat cultivars (Figure 3.30). In fact, Nusrat had a significantly higher K/Na ratio in all applications than Adana-99, which is the other bread wheat variety. Under control salinity conditions, moderate and severe B toxicity treatments significantly increased the straw K/Na ratio of Nusrat (Figure 3.30A).

Under medium salinity conditions, straw K/Na ratio of Nusrat tended to increase when moderate and severe B toxicity was applied (Figure 3.30B). In another bread wheat variety Adana-99, moderate B toxicity significantly increased straw K/Na ratio, but there was no difference observed between severe B toxicity and moderate B toxicity application.

Straw K/Na ratio significantly increased in the presence of B toxicity in the high salinity applied Nusrat cultivar (Figure 3.30C). Treatment of moderate and severe B toxicity tended to increase straw K/Na ratio in Adana-99.

Also, as can be seen from the differences in the y-axes of the figures, medium and high salinity stress gradually reduced straw K/Na ratio in all four wheat cultivars (Figure 3.30).

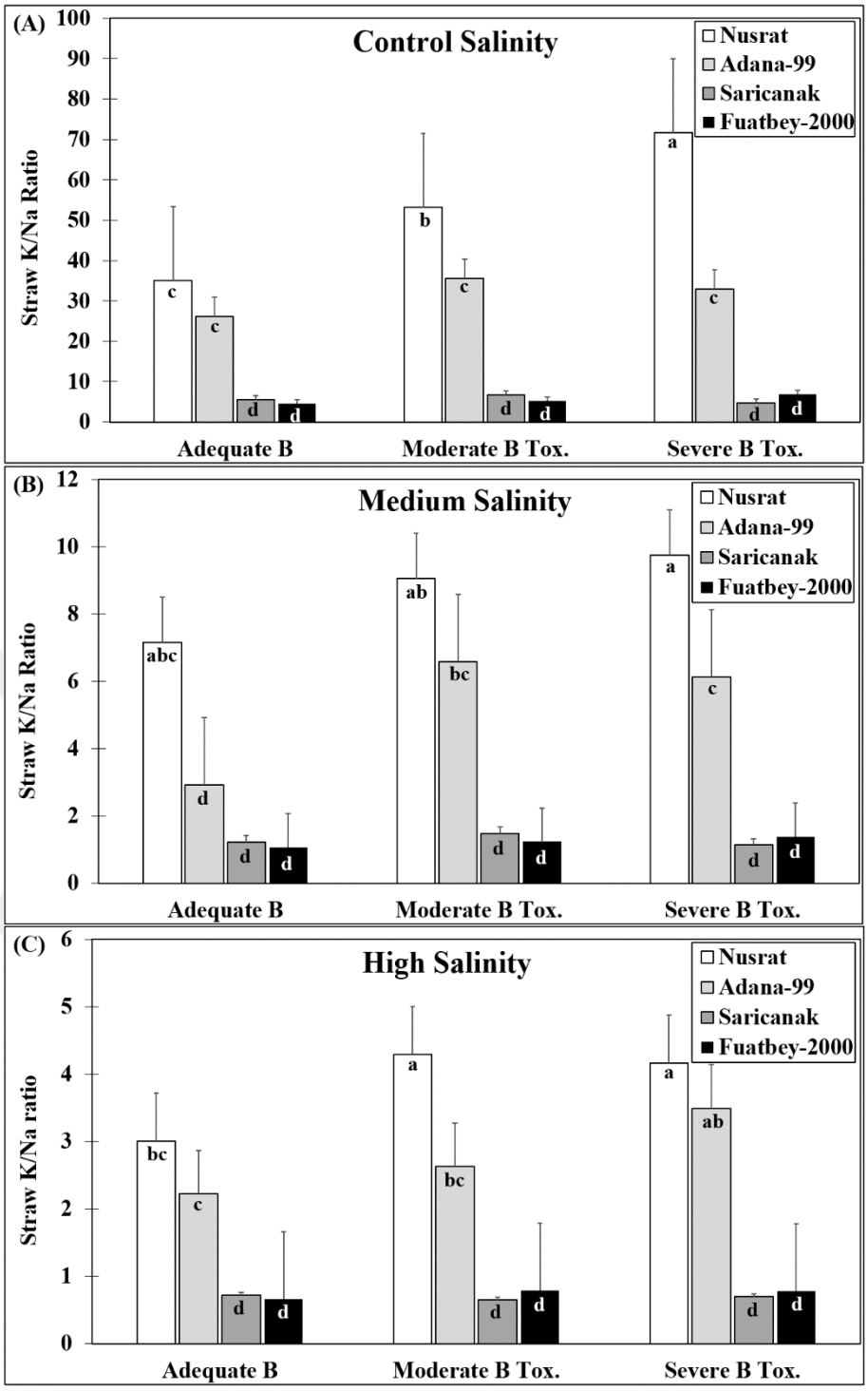

Figure 3.30: Effects of (A) control: 3.5 $dS.m^{-1}$ EC_e, (B) medium: 5.5 $dS.m^{-1}$ EC_e, (C) high: 7.5 $dS.m^{-1}$ EC_e treatments on relative straw K/Na ratios of completely mature bread wheat (*Triticum aestivum* cv. Nusrat and Adana-99) and durum wheat (*Triticum durum* cv. Saricanak and Fuatbey-2000) grown in soil at different levels of B supply (adequate: 0 $mg.kg^{-1}$ H_3BO_3; moderate: 10 $mg.kg^{-1}$ H_3BO_3; severe: 20 $mg.kg^{-1}$ H_3BO_3) under greenhouse conditions. Please note that scales of three graphs are different. Different letters indicate significant differences between means according to two-way ANOVA and Tukey's HSD test ($p \leq 0.05$).

Table 3.46: Three-way analysis of variance of the effects of salinity, B toxicity, and cultivar and their interaction on yield parameters of completely mature cultivars of bread wheat (*Triticum aestivum* cv. Nusrat and Adana-99) and durum wheat (*Triticum durum* cv. Saricanak and Fuatbey-2000) grown in soil under greenhouse conditions: degrees of freedom, F value probabilities, and Tukey's HSD test scores.

Source of Variation	**DF**	**Straw DW**		**Grain yield**		**Harvest index**	
		F Pr.	$HSD_{0.05}$	F Pr.	$HSD_{0.05}$	F Pr.	$HSD_{0.05}$
Soil Salinity (SS)	2	<0.001	0.1	<0.001	0.1	0.0013	2
B Supply (BS)	2	<0.001	0.1	<0.001	0.1	0.001	2
Cultivar (CV)	3	<0.001	0.2	<0.001	0.1	<0.001	2
SS*BS	4	0.0069	0.3	0.086	0.2	0.0083	4
BS*CV	6	0.2054	0.4	<0.001	0.3	<0.001	5
SS*CV	6	0.091	0.4	<0.001	0.3	<0.001	5
SS*BS*CV	12	0.258	0.8	<0.001	0.6	<0.001	11
Source of Variation	**DF**	**Spike number**		**Grain number**		**TGW**	
		F Pr.	$HSD_{0.05}$	F Pr.	$HSD_{0.05}$	F Pr.	$HSD_{0.05}$
Soil Salinity (SS)	2	<0.001	0.2	<0.001	18	0.0693	2
B Supply (BS)	2	<0.001	0.2	0.001	18	0.0101	2
Cultivar (CV)	3	<0.001	0.2	<0.001	23	<0.001	3
SS*BS	4	0.5766	0.5	0.0315	42	0.0058	5
BS*CV	6	0.0194	0.6	<0.001	47	0.0092	6
SS*CV	6	0.0129	0.6	0.1175	50	<0.001	6
SS*BS*CV	12	0.0896	1.2	0.0021	82	0.0086	13

Medium and high salinity treatment significantly reduced straw DW (Table 3.46; Table 3.47). Straw DW was not affected by moderate B toxicity application, while severe B toxicity treatment significantly reduced it. According to the ANOVA, it was determined that the salinity and B toxicity interaction significantly affected the straw DW (Table 3.46). The highest straw DW was detected in plants grown under salinity control, regardless of the B toxicity treatment. Among the four wheat cultivars, the highest straw DW was found in Fuatbey-2000. The other variety of durum wheat, Saricanak, had the lowest straw DW.

Medium and high soil salinity treatment significantly reduced the grain yield per plant in all four wheat cultivars except Fuatbey-2000 (Table 3.46; Table 3.47). The application of moderate B toxicity did not affect grain yield, but the treatment of severe B toxicity significantly decreased grain yield. Bread wheat cultivars were found to have a higher grain yield than durum wheat varieties. In addition, it was determined that the grain yield of Nusrat, a bread wheat variety, was 22% higher than the other bread wheat variety Adana-99.

Medium soil salinity application tended to decrease the harvest index (HI), while high salinity treatment significantly decreased it (Table 3.46; Table 3.47). The HI of wheat increased by 11% and 6% with the application of moderate and severe B toxicity, respectively. Harvest index of Nusrat and Adana-99 were found to be very similar. However, it was detected that the HI of Saricanak and Fuatbey-2000, which are durum wheat varieties, were approximately 42% and 138% lower than bread wheat varieties, respectively.

Table 3.47: Straw dry weight (DW), grain yield, and harvest index (HI) of completely mature bread wheat (*Triticum aestivum* cv. Nusrat and Adana-99) and durum wheat (*Triticum durum* cv. Saricanak and Fuatbey-2000) grown in soil at different levels of soil salinity stress (control: 3.5 $dS.m^{-1}$ EC_e; medium: 5.5 $dS.m^{-1}$ EC_e; high: 7.5 $dS.m^{-1}$ EC_e) and B supply (adequate: 0 $mg.kg^{-1}$ H_3BO_3; moderate: 10 $mg.kg^{-1}$ H_3BO_3; severe: 20 $mg.kg^{-1}$ H_3BO_3).

B Supply	Soil Salinity	Cultivar	Straw DW ($g.plant^{-1}$)	Grain yield ($g.plant^{-1}$)	Harvest index (%)
Adequate B	Control	Nusrat	3.0 ± 0.5	2.5 ± 0.2	38 ± 5
		Adana-99	2.5 ± 0.3	2.3 ± 0.3	40 ± 1
		Saricanak	1.7 ± 0.2	0.7 ± 0.3	25 ± 6
		Fuatbey-00	3.7 ± 0.3	0.5 ± 0.3	10 ± 5
	Medium	Nusrat	2.2 ± 0.3	1.6 ± 0.4	34 ± 8
		Adana-99	2.1 ± 0.3	1.4 ± 0.2	31 ± 3
		Saricanak	1.4 ± 0.2	0.7 ± 0.2	25 ± 4
		Fuatbey-00	3.2 ± 0.6	1.3 ± 0.1	24 ± 3
	High	Nusrat	2.1 ± 0.3	1.4 ± 0.5	32 ± 6
		Adana-99	1.9 ± 0.2	1.1 ± 0.1	29 ± 2
		Saricanak	1.4 ± 0.2	0.5 ± 0.1	20 ± 4
		Fuatbey-00	3.4 ± 0.3	1.4 ± 0.1	24 ± 1
Moderate B Toxicity	Control	Nusrat	2.5 ± 0.1	2.5 ± 0.2	41 ± 3
		Adana-99	2.3 ± 0.1	2.1 ± 0.2	39 ± 3
		Saricanak	1.5 ± 0.1	1.3 ± 0.2	36 ± 4
		Fuatbey-00	3.7 ± 0.1	1.1 ± 0.3	20 ± 5
	Medium	Nusrat	2.1 ± 0.2	2.2 ± 0.2	42 ± 3
		Adana-99	2.1 ± 0.2	1.7 ± 0.2	36 ± 4
		Saricanak	1.3 ± 0.2	0.8 ± 0.2	29 ± 2
		Fuatbey-00	4.1 ± 0.8	1.1 ± 0.1	17 ± 3
	High	Nusrat	2.0 ± 0.2	1.9 ± 0.2	41 ± 2
		Adana-99	1.6 ± 0.2	1.2 ± 0.1	35 ± 1
		Saricanak	1.1 ± 0.1	0.4 ± 0.1	19 ± 2
		Fuatbey-00	3.2 ± 0.2	0.7 ± 0.3	14 ± 6
Severe B Toxicity	Control	Nusrat	2.7 ± 0.2	2.2 ± 0.1	36 ± 1
		Adana-99	2.2 ± 0.1	2.1 ± 0.2	41 ± 3
		Saricanak	1.4 ± 0.1	1.0 ± 0.3	33 ± 5
		Fuatbey-00	3.8 ± 0.3	0.2 ± 0.1	4 ± 1
	Medium	Nusrat	2.0 ± 0.4	1.8 ± 0.2	39 ± 6
		Adana-99	1.8 ± 0.2	1.4 ± 0.4	38 ± 4
		Saricanak	1.1 ± 0.1	0.6 ± 0.1	26 ± 3
		Fuatbey-00	2.9 ± 0.5	0.8 ± 0.2	17 ± 3
	High	Nusrat	1.7 ± 0.4	1.4 ± 0.2	38 ± 5
		Adana-99	1.3 ± 0.2	1.1 ± 0.2	37 ± 3
		Saricanak	0.9 ± 0.1	0.6 ± 0.1	29 ± 3
		Fuatbey-00	2.7 ± 0.6	0.4 ± 0.1	11 ± 3

Straw DW $HSD_{0.05}$ (SS; BS; CV; SSxBS; SSxCV; BSxCV; SSxBSxCV)= (0.1; 0.1; 0.2; 0.3; *n.s.*; *n.s.*; *n.s.*)
Grain yield $HSD_{0.05}$ (SS; BS; CV; SSxBS; SSxCV; BSxCV; SSxBSxCV)= (0.1; 0.1; 0.1; *n.s.*; 0.3; 0.3; 0.6)
Harvest index $HSD_{0.05}$ (SS; BS; CV; SSxBS; SSxCV; BSxCV; SSxBSxCV)= (2; 2; 2; 4; 5; 5.; 11)

High salinity and severe B toxicity treatments significantly reduced the spike number (Table 3.46; Table 3.48). However, the spike number was not affected significantly by medium soil salinity and moderate B toxicity applications. It was determined that the durum wheat variety, Saricanak, had a significantly higher spike number than other cultivars (Table 3.46; Table 3.48).

Medium and high soil salinity treatments significantly reduced the grain number in all wheat varieties except the Fuatbey-2000 cultivar (Table 3.46; Table 3.48). In plants treated with moderate B toxicity, grain numbers did not change drastically but decreased significantly in plants treated with severe B toxicity. It was determined that the grain number was higher in bread wheat varieties than in durum wheat varieties. However, significant differences were observed between cultivars. The grain number of Nusrat was 14% higher than the other bread wheat cultivar Adana-99. Similarly, it was determined that the grain number of Fuatbey-2000 was significantly higher than the other durum wheat variety, Saricanak, and the difference between them was determined as 35% (Table 3.46; Table 3.48).

According to the ANOVA result, soil salinity did not affect thousand grain weight (TGW) significantly (Table 3.46; Table 3.48). The application of moderate B toxicity to plants significantly increased the TGW, while the application of severe B toxicity did not affect it. The TGWs of Nusrat and Adana-99, which are bread wheat varieties, were found to be quite similar. However, it was determined that the TGW of Saricanak was approximately 32% lower than the bread wheat varieties. The TGW of Fuatbey-2000, another durum wheat variety, was 61% lower than the bread wheat varieties (Table 3.46; Table 3.48).

Table 3.48: Spike number, grain number, and thousand grain weight (TGW) of completely mature bread wheat (*Triticum aestivum* cv. Nusrat and Adana-99) and durum wheat (*Triticum durum* cv. Saricanak and Fuatbey-2000) grown in soil at different levels of soil salinity stress (control: 3.5 $dS.m^{-1}$ EC_e; medium: 5.5 $dS.m^{-1}$ EC_e; high: 7.5 $dS.m^{-1}$ EC_e) and B supply (adequate: 0 $mg.kg^{-1}$ H_3BO_3; moderate: 10 $mg.kg^{-1}$ H_3BO_3; severe: 20 $mg.kg^{-1}$ H_3BO_3).

B Supply	Soil Salinity	Cultivar	Spike number ($plant^{-1}$)	Grain number ($plant^{-1}$)	Thousand grain weight (g)
Adequate B	Control	Nusrat	2.2 ± 0.6	319 ± 49	38 ± 1
		Adana-99	2.5 ± 0.3	324 ± 67	37 ± 4
		Saricanak	3.3 ± 0.7	126 ± 40	29 ± 8
		Fuatbey-00	1.9 ± 0.2	88 ± 31	27 ± 9
	Medium	Nusrat	2.4 ± 0.7	282 ± 64	30 ± 10
		Adana-99	2.7 ± 0.3	250 ± 44	27 ± 5
		Saricanak	2.8 ± 0.2	135 ± 24	27 ± 4
		Fuatbey-00	1.9 ± 0.3	215 ± 22	31 ± 3
	High	Nusrat	1.9 ± 0.3	167 ± 33	39 ± 3
		Adana-99	1.9 ± 0.3	153 ± 31	36 ± 6
		Saricanak	2.0 ± 0.4	82 ± 23	25 ± 2
		Fuatbey-00	1.9 ± 0.2	240 ± 20	28 ± 4
Moderate B Toxicity	Control	Nusrat	1.9 ± 0.4	297 ± 10	42 ± 3
		Adana-99	2.3 ± 0.3	303 ± 30	35 ± 5
		Saricanak	2.7 ± 0.6	185 ± 44	36 ± 3
		Fuatbey-00	1.7 ± 0.1	171 ± 39	31 ± 5
	Medium	Nusrat	1.6 ± 0.3	223 ± 5	48 ± 5
		Adana-99	2.0 ± 0.3	226 ± 23	37 ± 2
		Saricanak	2.9 ± 0.3	133 ± 19	31 ± 3
		Fuatbey-00	2.4 ± 0.5	180 ± 36	26 ± 4
	High	Nusrat	1.2 ± 0.2	220 ± 35	44 ± 4
		Adana-99	1.9 ± 0.4	161 ± 31	35 ± 1
		Saricanak	2.4 ± 0.6	87 ± 7	22 ± 2
		Fuatbey-00	1.9 ± 0.3	145 ± 60	21 ± 5
Severe B Toxicity	Control	Nusrat	1.8 ± 0.3	318 ± 31	35 ± 2
		Adana-99	2.3 ± 0.2	250 ± 65	45 ± 13
		Saricanak	2.1 ± 0.2	124 ± 12	34 ± 4
		Fuatbey-00	1.7 ± 0.5	83 ± 41	11 ± 2
	Medium	Nusrat	1.6 ± 0.7	212 ± 11	40 ± 5
		Adana-99	1.5 ± 0.3	169 ± 36	42 ± 4
		Saricanak	2.4 ± 0.4	113 ± 17	27 ± 2
		Fuatbey-00	2.2 ± 0.7	177 ± 52	22 ± 2
	High	Nusrat	1.5 ± 0.3	195 ± 49	36 ± 7
		Adana-99	1.4 ± 0.2	131 ± 24	40 ± 1
		Saricanak	2.0 ± 0.4	90 ± 28	28 ± 3
		Fuatbey-00	2.0 ± 0.5	146 ± 32	15 ± 2

Spike number $HSD_{0.05}$ (SS; BS; CV; SSxBS; SSxCV; BSxCV; SSxBSxCV)= (0.2; 0.2; 0.2; *n.s.*; 0.5; 0.5; *n.s.*)
Grain number $HSD_{0.05}$ (SS; BS; CV; SSxBS; SSxCV; BSxCV; SSxBSxCV)= (18; 18; 23 42; 48; *n.s.*; 107)
TGW $HSD_{0.05}$ (SS; BS; CV; SSxBS; SSxCV; BSxCV; SSxBSxCV)= (*n.s.*; 2; 3; 5; 6; 6; 13)

Salinity had no significant effect on grain B concentration (Table 3.44; Table 3.49). Moderate and severe B toxicity applications significantly increased the grain B concentration. The durum wheat variety Saricanak and the bread wheat variety Adana-99 had very similar grain B concentrations (Table 3.44; Table 3.49).

Grain Na concentration significantly increased with the application of medium and high soil salinity (Table 3.44; Table 3.49). Boron toxicity application did not significantly affect grain Na concentration. While moderate B toxicity application significantly decreased grain Na concentration, only a decrease trend was observed in the grain Na concentration of wheat with severe B toxicity treatment. It was determined that the grain Na concentration of bread wheat cultivars were significantly lower than the durum wheat varieties (Table 3.44; Table 3.49).

While the medium salinity decreased the grain K concentration, the high salinity treatment did not affect the grain K concentration (Table 3.44; Table 3.49). Severe B toxicity application increased grain K concentration by 14%, while moderate B toxicity had no significant effect. Bread wheat cultivars had significantly lower grain K concentration than durum wheat varieties. Besides, durum wheat varieties Fuatbey-2000 and Saricanak differed among themselves. Fuatbey-2000 had a 24% higher grain K concentration than Saricanak (Table 3.44; Table 3.49).

Table 3.49: Grain B, Na, and K concentrations of completely mature bread wheat (*Triticum aestivum* cv. Nusrat and Adana-99) and durum wheat (*Triticum durum* cv. Saricanak and Fuatbey-2000) grown in soil at different levels of soil salinity stress (control: 3.5 $dS.m^{-1}$ EC_e; medium: 5.5 $dS.m^{-1}$ EC_e; high: 7.5 $dS.m^{-1}$ EC_e) and B supply (adequate: 0 $mg.kg^{-1}$ H_3BO_3; moderate: 10 $mg.kg^{-1}$ H_3BO_3; severe: 20 $mg.kg^{-1}$ H_3BO_3).

B Supply	Soil Salinity	Cultivar	Grain Concentration		
			B ($mg.kg^{-1}$)	Na (mg.kg-1)	K (%)
Adequate B	Control	Nusrat	1.4 ± 0.3	36 ± 7	0.55 ± 0.05
		Adana-99	1.7 ± 0.2	21 ± 3	0.54 ± 0.04
		Saricanak	1.7 ± 0.2	25 ± 11	0.65 ± 0.07
		Fuatbey-00	1.8 ± 0.4	43 ± 3	0.82 ± 0.06
	Medium	Nusrat	1.4 ± 0.2	109 ± 44	0.60 ± 0.07
		Adana-99	1.4 ± 0.3	117 ± 63	0.55 ± 0.05
		Saricanak	1.4 ± 0.2	80 ± 54	0.59 ± 0.02
		Fuatbey-00	1.5 ± 0.1	199 ± 38	0.72 ± 0.07
	High	Nusrat	1.5 ± 0.1	128 ± 60	0.51 ± 0.02
		Adana-99	2.3 ± 1.0	20 ± 16	0.52 ± 0.03
		Saricanak	1.9 ± 0.7	607 ± 154	0.75 ± 0.18
		Fuatbey-00	1.7 ± 0.4	379 ± 36	0.75 ± 0.03
Moderate B Toxicity	Control	Nusrat	2.8 ± 0.4	31 ± 7	0.56 ± 0.04
		Adana-99	4.0 ± 0.6	29 ± 13	0.56 ± 0.03
		Saricanak	3.8 ± 0.5	25 ± 3	0.61 ± 0.04
		Fuatbey-00	3.3 ± 0.1	57 ± 9	0.79 ± 0.05
	Medium	Nusrat	2.6 ± 0.6	60 ± 37	0.56 ± 0.04
		Adana-99	3.8 ± 0.2	40 ± 30	0.55 ± 0.02
		Saricanak	4.0 ± 1.6	68 ± 31	0.66 ± 0.04
		Fuatbey-00	3.2 ± 0.5	122 ± 24	0.73 ± 0.02
	High	Nusrat	2.8 ± 0.6	106 ± 32	0.53 ± 0.02
		Adana-99	4.3 ± 0.5	36 ± 13	0.57 ± 0.03
		Saricanak	5.2 ± 1.0	307 ± 81	0.72 ± 0.06
		Fuatbey-00	3.4 ± 0.7	410 ± 120	0.92 ± 0.12
Severe B Toxicity	Control	Nusrat	5.1 ± 0.3	57 ± 37	0.68 ± 0.07
		Adana-99	6.4 ± 0.8	32 ± 13	0.58 ± 0.03
		Saricanak	7.3 ± 1.0	18 ± 5	0.61 ± 0.03
		Fuatbey-00	7.2 ± 1.5	80 ± 18	1.12 ± 0.08
	Medium	Nusrat	5.1 ± 0.6	92 ± 55	0.63 ± 0.08
		Adana-99	9.2 ± 2.5	23 ± 6	0.56 ± 0.02
		Saricanak	8.9 ± 2.0	116 ± 97	0.64 ± 0.05
		Fuatbey-00	4.8 ± 0.5	141 ± 28	0.94 ± 0.04
	High	Nusrat	5.4 ± 1.1	174 ± 122	0.62 ± 0.06
		Adana-99	8.1 ± 0.7	24 ± 2	0.56 ± 0.01
		Saricanak	7.8 ± 1.1	296 ± 256	0.70 ± 0.04
		Fuatbey-00	5.9 ± 0.9	369 ± 221	0.93 ± 0.04

Spike number $HSD_{0.05}$ (SS; BS; CV; SSxBS; SSxCV; BSxCV; SSxBSxCV)= (0.2; 0.2; 0.2; *n.s.*; 0.5; 0.5; *n.s.*)
Grain number $HSD_{0.05}$ (SS; BS; CV; SSxBS; SSxCV; BSxCV; SSxBSxCV)= (18; 18; 23 42; 48; *n.s.*; 107)
TGW $HSD_{0.05}$ (SS; BS; CV; SSxBS; SSxCV; BSxCV; SSxBSxCV)= (*n.s.*; 2; 3; 5; 6; 6; 13)

4. DISCUSSION

Combined stress factors on plants become more common with the impact of changing environmental conditions and climate change. Under the influence of these ecological factors, simultaneously occurring drought and heat stress became the most obvious stress combination [Prasad et al., 2011]. Stress factors such as salinity, drought, heat, high light, oxidative stress, mineral toxicity, and nutrient deficiency would eventually interact with each other in salt-affected soil on a hot, dry, and sunny summer day.

In the literature, there are only a few studies examining plant responses to the co-occurrence of salinity and B toxicity. Interaction of these stressors sometimes caused more severe results [Holloway and Alston, 1992], [Bastías et al., 2004], [Yermiyahu et al., 2008] on plants than individual stress factors, but some studies reported alleviating effects of one stress on another one [Alpaslan and Gunes, 2001]. Apart from this antagonistic and synergistic effects, some researchers showed that co-occurring stress factors may not have any effect on each other [Bingham et al., 1987], [Edelstein et al., 2005]. In the literature, there are studies showing the effect of salinity and B toxicity relationship on different wheat cultivars [Naz et al., 2018], [Wimmer and Goldbach, 2012]. However, there was no consensus among these studies. Therefore, in this study, different levels of salinity and B toxicity were applied to bread and durum wheat cultivars grown under hydroponic and soil conditions. As a result of the experiments, it was aimed to elucidate the effects of salinity and B toxicity on wheat growth, mineral distribution in various parts of the shoot and root, stress-related physiological parameters, oxidative stress markers, and yield.

4.1. Hydroponic experiments

The effects of salinity and B toxicity on plant growth varies according to plant species and genotype. Such differences can be explained by genotype-specific tolerance mechanisms. In this experiment, the shoot DW was drastically reduced by salinity stress, while B toxicity and salinity-B toxicity combination did not affect shoot biomass (Table 3.2; Table 3.8; Table 3.13). Due to the adaptation of various plants to different climatic and environmental conditions, it may be normal for the plant species exposed to the interaction of salinity*B toxicity to affect biomass differently [Alpaslan and Gunes, 2001], [Ben-Gal and Shani, 2002], [Díaz and Grattan, 2009]. This situation can even be seen in different cultivars of the same type of plants [Holloway and Alston, 1992], [Bingham et al., 1987]. While the effect of boron toxicity application was observed as necrosis affecting leaf tips and margins (Figure 3.1; Figure 3.5; Figure 3.10; Figure 3.11), dry weight was not affected (Table 3.2; Table 3.8; Table 3.13). Grieve and Poss (2000) described that the shoot biomass of wheat was reduced by salinity to a greater degree than by B. The shoot dry weight of plants treated with B toxicity may differ according to the species and cultivars [Masood et al., 2012], [Eraslan et al., 2007] [Marcar et al., 1999]. According to the results obtained from the third experiment in hydroponic media, the shoot biomass of durum wheat is 30% higher than that of bread wheat (Table 3.13).

In conformity with the results reported in this study, in the literature [Kumar et al., 2015], [Gunes et al., 2006], it has been shown that B toxicity significantly decreased root biomass of different plant species and wheat (Table 3.2; Table 3.9; Table 3.14). Salinity alone or in combination with boron toxicity did not have a positive or negative effect on root dry weight. In contrast, root growth was impaired by B toxicity alone [Naz et al., 2018], [Turan et al., 2009]. However, according to the just first hydroponic experiment, the root dry weight of the plants without toxic B but only treated with high salt tended to decrease (Table 3.2). It was noted that the amount of root dry weight differed between species: bread wheat 28% higher than durum wheat (Table 3.2). This result contrasts with the shoot dry weight result.

When the element concentrations were measured, it was observed that the application of B toxicity increased the shoot B concentration and content (Figure 3.3; Table 3.10; Table 3.15). It is inevitable that plants with excess B in their growth

medium will translocate excess B from root to shoot. The phenomenon that makes this compulsory can be explained as follows: the B micronutrient is immobile in most plants and hence is mainly transported through xylem, which depends on the transpiration stream [Brown et al., 2002], [Nable et al., 1997]. One of the most important tolerance mechanisms developed by plants against salt stress is stomatal closure and therefore a decrease in transpiration [Alpaslan and Gunes, 2001], [Bastías et al., 2004]. As can be observed as a consequence of the third hydroponic experiment, transpiration of both bread and durum wheat species decreased under medium and high salinity stress (Table 3.21; Figure 3.13). When both salt and toxic boron were added to the growth medium, especially when high doses were applied, it was determined that shoot boron concentration and content decreased (Figure 3.3; Table 3.10; Table 3.15). The greatest phenotypic indicator of toxic B, which is transferred to the shoot by transpiration and accumulates on the tips and margins of mature leaves, is the formation of necrosis in the plant parts where it is carried [Stangoulis and Reid, 2002], [Poss et al., 1998]. Just as the B concentration and content decreased in the presence of medium and high salinity, the leaf area affected by necrosis also declined (Figure 3.5; Figure 3.7). These results led us to question whether there is a direct relationship between transpiration and severity of necrosis. The answer to the question of whether the necrosis severity is related to the transpiration of the plant lies below (Table 4.1). Transpiration and B content are highly correlated with each other in both wheat species (Figure 4.1.A; Figure 4.1.C). Therefore, a plant that exposed to salinity and B toxicity stresses at the same time, reduces the transpiration flow and shoot B translocation (content and concentration), which alleviates the necrotic symptoms seen on the mature leaf tips and margins (Figure 4.1.B; Figure 4.1.D).

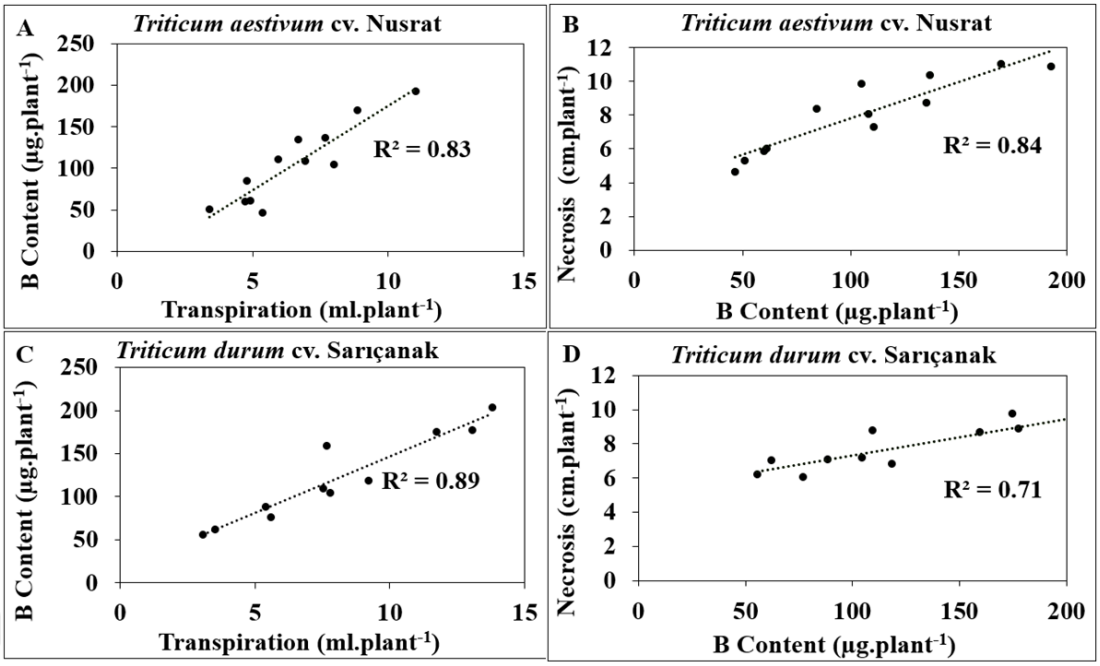

Figure 4.1: Relationship between (A) transpiration rate and B content, (B) B content and necrosis severity of 4-weeks-old bread wheat (*Triticum aestivum* cv. Nusrat) and (C) transpiration rate and B content (D) B content and necrosis severity of durum wheat (*Triticum durum* cv. Saricanak) grown hydroponically at different levels of salinity (control: 0 mM NaCl; medium: 60 mM NaCl; high 120 mM NaCl) and B supply (control: 2 µM; toxic: 1 mM).

As mentioned in the previous sections, various toxicity doses were tested in the first hydroponic experiment. Appropriate salinity and B supply doses were used in the second and third hydroponic experiments. The moderate dose (1 mM) of B toxicity applied in the first hydroponic experiment and the severe B toxicity dose (1 mM) used in the 2nd and 3rd experiments are equal. When B was used at a range of 0.5-1 mM, the toxicity was not found to be very severe, but when applied as 2 mM and beyond, it had very severe effects [Bastías et al., 2004], [Princi et al., 2015]. Consequently, the application of severe B toxicity in experiments 2 and 3, of course, caused stress in plants (Table 3.10; Table 3.15). For this reason, since the shoot biomass does not decrease importantly and the plants are not shocked, the antagonistic effects triggered by the combination of salinity and B toxicity come to the fore and a decline in root B concentration is observed. The same situation was noticed in hydroponic experiment 1 and the concentration of root B in plants treated with moderate B toxicity decreased with the application of salinity (Table 3.3). However, when severe B toxicity (2.5 mM) applied in the same experiment, the biomass of the plants decreased drastically, and the concentration effect observed.

Boron accumulated in the older shoot tissues and shoot tips and younger parts usually have very low B. However, roots directly exposed to severe B. Therefore, root growth was inhibited by B toxicity (Princi et al., 2016). This situation also same in Al toxicity. The most easily affected location of Al toxicity is the root in plant. Root elongations a process of cell division, but Al toxicity block the mechanism of cell division. Under Al toxicity, root growth inhibited but shoot growth was not affected by Al toxicity (Panda et al., 2009).

Low, medium, and high salinity stress decreased shoot K concentration and increased shoot Na concentration in bread and durum wheat (Table 3.4; Table 3.5; Table 3.10; Table 3.16; Table 3.17). Durum wheat (AB genomes) is more susceptible to salinity than bread wheat (ABD genomes), a trait that limits its expansion to fields with saline soils [Munns et al., 2000]. Salinity tolerance in bread wheat is associated with a locus on the D genome that results in low Na^+ uptake and increased K/Na discrimination. Elevated Na concentration can inhibit K uptake due to ionic stress, which is a critical component of salt stress [Grattan and Grieve, 1998], [Navarro et al., 2000]. The primary reason for K^+ reduction due to excess Na^+ caused by salinity is the influx of competition between these two cationic nutrients. As a natural consequence of this competition, the K/Na ratio decreased (Table 3.6; Table 3.11; Table 3.18) in shoot and root parts of plants grown in saline media [Masood et al., 2012], [Naz et al., 2018]. It is also known that the severity of the reducing effect of salt on shoot K concentration depends on the plant species [Wang and Wu, 2013], [Läuchli and Grattan, 2007]. As evidence to support this information, when salt stress is applied, it was determined that the K concentration of Saricanak decreases more than that of bread wheat cultivar Nusrat (Table 3.17). Higher salinity tolerance in bread wheat compared with durum wheat has been linked to its improved ability to exclude Na^+ from uptake. It seems that the exclusion of Na^+ alone is not always sufficient to improve the resistance of plant salinity, and other physiological characteristics should also be considered [Cuin et.al., 2008]. K^+ homeostasis have a critical role in plant salinity tolerance. Mechanism used by bread wheat, namely more superior K^+ retention ability in the leaf mesophyll, also contributes to this distinction [Wu et al., 2014].

Boron toxicity alone or in its interaction with salinity did not reduce the shoot K concentration and content (Table 3.5, Table 3.10, Table 3.17). Similar results were obtained in the experiments conducted with various plant species in the literature

[Mohamed et al., 2016], [Ben-Gal and Shani, 2002]. Since B toxicity did not significantly affect both Na and K concentrations, the K/Na ratio was not affected by B toxicity. Therefore, we can say that the K/Na ratio decreases only under salinity due to ionic competition (Table 3.6).

It is known that the partitioning of mineral nutrients in plants is affected by numerous factors [Wang and Below, 1998], [Marschner, 2011]. Excess B is transported to leaf tips and margins and causes necrotic formations [Sutton et al., 2007]. Therefore, it is natural that the highest B concentration in the tissue is detected in the necrotic parts of older leaf tips (Table 3.12). The reducing effect of salinity on B concentration was also observed in the necrotic parts of older leaves. It was determined that salinity increases Na concentration in all shoot tissues, and Na accumulation is less in young leaves than in old leaves. While the lowest K concentration is naturally observed in necrotic tissues, the K-reducing effect of salinity was found mostly in non-necrotic parts of old leaves [Blumwald, 2000]. The explanation for this situation that Na is the dominant cation in the relevant tissue part.

It was observed that salinity also significantly enhanced the root Na concentration and content (Table 3.4, Table 3.16). In the saline condition, the application of B toxicity did not affect the Na concentration of the roots but decreased the root Na content [Martinez-Ballesta et al., 2008]. The answer for this circumstance can be identified as the 'concentration-effect' that occurs as a result of the toxic level of B disrupting the plant's root tissues and thus restricting root growth [Marschner, 2011]. The adverse effects of salinity on the root K concentration and content were determined (Table 3.5, Table 3.10). Like the shoot K/Na ratio, the root K/Na ratio also displayed a significant decline in the presence of salinity (Table 3.6, Table 2.4, Table 3.7).

Calcium, as a macronutrient, has key functions in plants such as cell membrane integrity [Hepler, 2005], selective ion absorption [Marschner, 2011], cell wall stability [Yamauchi et al., 1986], cell elongation [Burstrom, 1968], cell division [Rasmussen, 1970], and pollen tube formation [Kwack, 1967], which are, like B, influencing physiological and biochemical processes [Turan et al., 2009]. It is a well-known principle that salinity reduces Ca concentration and content in both shoot and root [Rengel, 1992]. Data supporting this phenomenon can also be observed in our experiment results (Table 3.7, Table 3.10, Table 3.17). The reason for the decrease in

Ca concentration is the loss of function of aquaporins [Cabañero et al., 2006] due to the severe increase of Na ions or the decrease in aquaporin expression as a result of Ca starvation caused by salinity stress [Bastías et al., 2010].

The reduction in Mg concentration caused by salinity stress in both shoot and root tissues is probably the negative impact of excess Na^+ on the absorption of Mg^{2+} [Martínez-Ballesta et al., 2006], [Ferreyra et al., 1997], [Grieve et al., 2010]. Moreover, the Mg concentration in the tissues was similarly decreased by boric acid (H_3BO_3), which is the transport and deposition form of B in plants [Martinez-Ballesta et al., 2008]. The effect of salinity and B toxicity on the Mg concentration in root tissue draws attention to a different circumstance (Table 3.17). Boron toxicity triggered a decline in root Mg concentration in wheat grown in non-saline, low-saline, and medium-saline conditions, whereas B toxicity did not affect root Mg concentration in wheat grown in high-saline media. For this reason, salinity can be considered as the primary factor responsible for the reduction of Mg concentration in root tissues.

We assessed the influence of the relationship between salinity and B toxicity on another macronutrient, P. While no effect of these two stress agents on shoot P concentration was recorded, it was determined that they independently influence the concentration of shoot P (Table 3.17). In this experiment, salinity increased the shoot P concentration in both species. The effect of salinity on shoot P concentration may differ depending on many parameters. These may be plant species, cultivar, salt content, salt level, media type (soil or hydroponic), P concentration and form in the substrate [Grattan and Grieve, 1998]. In most cases, shoot P concentration is diminished by salinity [Sharpley et al., 1992], [Martinez and Lächli, 1991]. However, it was found that the shoot P concentration increased in the experiment conducted in a hydroponic growth medium using wheat plants [Masood et al., 2012]. On the other hand, B toxicity tended to decrease the concentration of shoot P but had no major impact.

Shoot Fe concentration was not affected by salinity and B toxicity interaction (Table 3.14). Depending on salinity composition, salinity level, and plant species, the impact of salinity stress on shoot Fe concentration differs markedly [Grattan and Grieve, 1998], [Sotiropoulos et al., 2006], [Bastías et al., 2010]. Shoot Fe concentration increased in wheat with medium salinity treatment (Table 3.19). In all conditions, the bread wheat variety was found to have a higher shoot Fe

concentration than the durum wheat cultivar. Furthermore, it was observed that the application of B toxicity reduced the shoot Fe concentration.

Both salinity and B toxicity reduced the shoot Zn concentration and aggravated each other's effects (Table 3.14, Table 3.19). In this regard, contradictions have been observed in the literature, and in the study conducted with cherry, it was determined that the interaction of salinity and B toxicity did not have a synergistic or antagonistic effect on the Zn concentration [Sotiropoulos et al., 2006]. In addition, there were studies that showed that Zn concentration increases with the application of B toxicity [Martinez-Ballesta et al., 2008]. Furthermore, in literature, salinity alone has been shown to increase [Ruiz et al., 1997] and decrease [Al-Harbi, 1995] Zn concentration [Grattan and Grieve, 1998].

Dose-dependent changes were observed in the effect of salinity on shoot Mn concentration. Compared to the control, it was determined that while medium salinity (60 mM) treatment increased shoot Mn concentration in both wheat varieties, high salinity (120 mM) application decreased Mn concentration (Table 3.14, Table 3.19). Several studies in the literature indicate that salinity reduces the concentration of shoot Mn [Cramer and Nowak, 1992], [Izzo et al., 1993]. It has also been found that B toxicity alone has a reducing effect on cherry Mn concentration [Sotiropoulos et al., 2006]. In our study, consistent with the literature, it was found that bread wheat has higher Mn concentration than durum wheat [Grattan and Grieve, 1998], [Bastías et al., 2010].

No significant effect of salinity and B toxicity interaction on shoot Cu concentration was detected (Table 3.14). As seen in the studies performed, the change in shoot Cu concentration varies depending on the source of salinity and the plant species [Rahman et al., 1993], [Izzo et al., 1993], [Grattan and Grieve, 1998].

Reactive oxygen species (ROS) including mainly H_2O_2 (hydrogen peroxide), $^1O_2$ (singlet oxygen), $O_2^{\cdot -}$ (superoxide radical), and $OH^{\cdot}$ (hydroxyl radical) cause cellular degradation, oxidative damage, and generally accumulate rapidly under abiotic stress conditions in organelles such as peroxisomes, chloroplasts, and mitochondria of plants [Mittler, 2002], [Gill and Tuteja, 2010].

Lipid peroxidation caused by ROS in all living organisms is considered a marker of stress [Mittler, 2002]. As a marker of lipid peroxidation and as a kind of measurement, MDA concentration increases (Table 3.22) when salinity stress is applied to plants [Pan et al., 2006], [Eraslan et al., 2007], [Meloni et al., 2003].

Because the MDA interferes with Fe, MDA assays are not very specific assays for analyzing the extent of lipid peroxidation (Sinha and Saxena, 2006). Under our control conditions, plants have a high Fe concentration (Table 3.19). This high Fe interferes with MDA and therefore control plants seem to have some MDA concentration. Also, some studies reported that B toxicity increases MDA concentration [Sang et al., 2015], [Cervilla et al., 2007]. However, in our experiments, it was determined that MDA concentration was not affected by B toxicity (Table 3.22), in accordance with the literature [Eraslan et al., 2007], [Hamurcu et al., 2016]. Why did B toxicity not increase MDA concentration? It is known that toxic B accumulates especially at the tips and margins of mature leaves [Nable et al., 1997]. Plants carry excess B to leaf tips and margins to minimize photosynthetic area loss in their leaves. Thus, the accumulation of B in the non-necrotic parts of the leaves is minimized and it can be speculated that this minimizes the oxidative effects of B toxicity on non-necrotic tissues. Since we expect to observe the increase of lipid peroxidation only in living tissue, the MDA concentration is not significantly affected by the application of B toxicity (Kayihan et al., 2017).

Relative electrolyte leakage (REL), which is another stress indicator, reflects the damage in cellular membranes caused by ROS [Qadir and Schubert, 2002]. The increase in REL indicates that membrane permeability is excessively increased, and membrane integrity is impaired [Zhu et al., 2004], [Blum and Ebercon, 1981]. It is known in the literature that both salinity and B toxicity increase REL [Bastías et al., 2010], [Karabal et al., 2003], [Tavallali et al., 2017]. In our experiment conducted on both bread and durum wheat species, we demonstrated that salinity and B toxicity significantly increased REL, in accordance with the literature (Table 3.20; Table 3.22). However, we would like to clarify the following question in order to avoid any question marks: Why did B toxicity enhance REL while not increasing the concentration of MDA? The answer to this question lies in the procedure for REL measurement. REL was measured from the third oldest leaf of 30-day-old plants, without removing necrotic parts. The integrity of the membrane is usually completely lost in the necrotic tissue, which is the tip and edge of the leaf, so electrolytes leak out. For this reason, we see the impact of B toxicity on REL.

It is necessary for the plant to control the ROS level with antioxidative enzymes (SOD, GR, APX, CAT) in order to cope with oxidative stress [Gill and

Tuteja, 2010], [Mittler, 2002]. Salinity increased SOD enzyme activity in both wheat species under both control and toxic boron conditions (Table 3.14). This result is consistent with the literature [Eraslan et al., 2007], [Tavallali et al., 2017]. In addition, it was determined that bread wheat SOD enzyme activity was higher than durum wheat (Table 3.14). It is known that SOD enzyme activity may vary among plant species [Naz et al., 2019]. There are studies in the literature suggesting that the application of B toxicity enhances or does not affect the SOD enzyme activity depending on the dose and the applied plant species [Ardic et al., 2009], [Princi et al., 2015], [Eraslan et al., 2007].

Although the activity of the GR enzyme in durum wheat was increased by salinity stress, it tended to decrease in bread wheat (Table 3.24). It has been shown in the literature that the effect of salinity on GR enzyme activity may differ between wheat cultivars and plant species [Sairam et al., 2005], [Hernández et al., 2000]. In this study, we found that B toxicity did not have a significant effect (Table 3.20) on GR enzyme activity [Masood et al., 2012].

High salt application significantly increased Saricanak APX enzyme activity (Table 3.20; Table 3.24). It was observed that APX enzyme activity was increased in Nusrat cultivar with medium salinity. Studies showing that the effect of salinity stress on the APX enzyme differ depending on the species are found in the literature [Hernández et al., 2000], [Tavallali et al., 2017]. While high salinity significantly increased the CAT activity in Nusrat, the same effect was not observed in Saricanak (Table 3.20; Table 3.24). In the literature, salinity was reported to increase the CAT enzyme activity [Tavallali et al., 2017], [Eraslan et al., 2007].

The specific (sp.) enzyme activity was expressed as activity per mg of Bradford protein [Hamurcu et al., 2016]. There was no significant effect of salinity and salt-B toxicity interaction on sp. SOD activity (Table 3.20). However, B toxicity diminished the sp. SOD activity of Nusrat in all three salinity treatments while it decreased the sp. SOD activity of Saricanak only under the non-saline conditions (Table 3.25). The increase in total protein concentration in plants treated with B toxicity can be considered as the main reason for this situation. In a study conducted on *P. distans*, it was found that the application of B toxicity usually diminished sp. SOD activity, although it differed by dosage [Hamurcu et al., 2016]. However, it was observed that the application of B toxicity increased the sp. SOD activity of *S.lycopersicum* [Cervilla et al., 2007]. In a paper related to wheat, no increase in

specific SOD activity was detected in plants treated with 0.5, 1, and 1.5 mM B toxicity [Naz et al., 2018].

It was determined that salinity and B toxicity applications tended to reduce the sp. GR enzyme activity of Nusrat, albeit independently (Table 3.20; Table 3.25). There was no consistent result in Saricanak related to sp. GR activity. It was observed that sp. GR activity decreased in various wheat cultivars with high salinity application [Sairam et al., 2005]. In addition, a decrease in GR activity was detected in *P.distans* treated with B toxicity [Hamurcu et al., 2016].

Specific APX and sp. CAT activity tended to decrease with the application of B toxicity (Table 3.25). The explanation for this may be the increase in the total protein concentration of the plants treated with B toxicity. Also, sp. GR and sp. CAT activities of *Pistacia vera* increased and decreased according to the severity of salinity and B toxicity treatments [Tavallali et al., 2017]. In addition, it was observed that the cultivar differences in wheat can show different results (Table 3.25) on sp. CAT activity [Naz et al., 2018].

Salinity stress restricts water uptake by reducing the water potential (Ψ_W) and causes water deficit in plants [Taiz and Zeiger, 2010]. Osmotic adjustment (osmoprotection) is necessary for the plants to maintain water uptake under stress conditions [Girma and Krieg, 1992]. Osmotic adjustment is the accumulation of solutes in cells in order to decrease the solute potential (Ψ_S) and thus enable continue water absorption at low water availability [Marschner, 2011], [Agarwal and Pandey, 2004]. Compatible solutes are of neutral or zwitterionic character and do not disturb the hydration shells of biological molecules [Hare and Cress, 1997]. Important compatible solutes are proline, glycine betaine, pinitol, trehalose, mannitol, and sorbitol [Demiral and Türkan, 2004]. In addition to osmotic adjustment, compatible solute accumulation is effective in ROS scavenging and membrane stabilization in higher plants [Mansour, 1998], [Munns and Tester, 2008].

Proline and glycine betaine (GB) concentrations were elevated in both wheat species in the presence of salinity stress and increased stress intensity (Table 3.20, Table 3.23). It was determined that the proline or GB concentration was not enhanced by B toxicity alone (Table 3.20). For this reason, we can say that salinity causes osmotic stress and thus increases the concentration of compatible solutes in the shoot of the plants as a stress response [Eraslan et al., 2007], [Agarwal and Pandey, 2004], [Farooq et al., 2015b], [Flowers and Colmer, 2008]. However, since

B toxicity did not cause possible osmotic stress, no significant increase was found in the proline and glycine betaine concentrations of the plants treated with only B toxicity [Karabal et al., 2003], [Yokoi et al., 2002], [Demiral and Türkan, 2004].

4.2. Soil experiments

Medium and high salinity significantly reduced germination percentage in both wheat varieties (Table 3.26, Figure 3.15; Table 3.27). There are also reports in the literature demonstrating the negative effect of salinity on germination percentage [Bonilla et al., 2005], [Francois et al., 1986], [Ismail, 2004]. Reduced water uptake of seeds due to the low osmotic potential of the soil [Akbarimoghaddam et al., 2011], changes in enzyme metabolism due to ion toxicity [Gomes-Filho et al., 2008], and loss of hormone balance [Khan et al., 2012] are the key reasons that salinity limits seed germination [Parihar et al., 2015]. Salinity stress delays germination (Figure 3.15), even though the final percentage of germinated seeds in medium salinity treatments would eventually be nearly the same with non-saline [Läuchli and Grattan, 2007]. However, further increases in salinity will ultimately decrease the percentage of germinated seeds as well.

It was observed that the application of B toxicity alone did not significantly affect the seed germination percentage (Table 3.30, Table 3.31) but only tended to decrease it probably because the plant roots had not yet developed enough to reach the toxic B in the soil [Yau and Ryan, 2008]. However, the interaction of salinity and B toxicity is thought to have a negative effect on the germination percentage [Mohamed et al., 2016], [Wimmer and Goldbach, 2012]. As can be seen in Table 3.31 as a result of the germination percentage measured 14 DAS, this phenomenon is consistent with our results.

The medium and high doses of salinity stress and severe B toxicity (30 ppm) significantly reduced plant height 7, 14, and 21 days after sowing (DAS) (Table 3.27). As a result of the application of salinity and B toxicity, it has been determined that the plant growth and hence the height decline in wheat and different plant species [Francois et al., 1986], [Parihar et al., 2015], [Kalayci et al., 1998], [Akbarimoghaddam et al., 2011].

Main effects of salinity and B toxicity and their combination decreased the shoot FW (Table 3.26, Figure 3.16). This was also observed in other studies

conducted before on wheat and different plant species [Parihar et al., 2015], [Mohamed et al., 2016], [Ben-Gal and Shani, 2003].

Shoot DW significantly reduced in wheat treated with medium (6 $dS.m^{-1}$) and high (8.5 $dS.m^{-1}$) soil salinity compared to non-saline (3.5 $dS.m^{-1}$) plants. It is a known fact that plant growth and DW are adversely impacted by salinity [Masood et al., 2012], [Del Carmen Rodríguez-Hernández et al., 2013]. According to the first soil experiment results, the shoot DW only tended to decrease as a result of mild (10 ppm) and moderate (20 ppm) B toxicity application while it was significantly reduced under severe (30 ppm) B toxicity (Figure 3.17). In the second soil experiment, similar findings were obtained, and it was found that applying both moderate (15 ppm) and severe (20 ppm) B toxicity significantly reduced the shoot DW. In the literature, it was observed that the application of 25 ppm B toxicity to soil reduced the shoot DW by 25-50% depending on the wheat species [Kalayci et al., 1998]. It was observed that main effect of B toxicity application in various plant species grown under soil conditions decreased the shoot DW [Aftab et al., 2015], [Ben-Gal and Shani, 2003], [Karimi and Tavallali, 2017].

It was observed that salinity and B toxicity stress factors decreased the tissue water content both separately and by aggravating each other's effect (Table 3.28). The impact of the stress combination on reducing the water content was also seen in studies conducted on rice [Farooq et al., 2015a] and pistachio [Karimi and Tavallali, 2017]. As a result of osmotic stress, which is one of the detrimental consequences of salinity, shoot water content reduces [Masood et al., 2012]. Reduction of water leads to loss of turgor and thus restriction in water availability [Díaz et al., 2011]. In addition, the toxic effect of B aggravates the decline of water transmission [Escobar et al., 2014].

In the first and second experiments established in the soil, all B toxicity applications, except adequate, significantly increased the shoot B accumulation (Table 3.29, Table 3.33). Shoot B concentration and content results in soil experiments were highly correlated with hydroponic experiment results. The antagonistic relationship between salinity and B toxicity on shoot B concentration and content was determined in both hydroponic and soil culture experiments in this thesis study (Figure 3.19, Figure 3.21). Similar results were observed in experiments conducted with tomato [Alpaslan and Gunes, 2001], cucumber and sorghum [Ismail, 2004] plants. It is thought that the concentration of shoot B decreases as the

transpiration rate diminishes with salinity [Bastías et al., 2004]. In addition, it is assumed that soil salinity decreases root development and therefore reduces root surface area and inhibits B uptake [Naz et al., 2019]. In the second soil experiment conducted with two different wheat species and 4 different cultivars, durum wheat varieties accumulated a higher amount of B in shoot than bread wheat cultivars (Table 3.33, Figure 3.20, Figure 3.22). Differences between wheat cultivars in terms of shoot B accumulation have been documented in various studies [Naz et al., 2019], [Wimmer and Goldbach, 2012].

Shoot Na concentration and content significantly increased in plants grown in salinity applied soils (Table 3.29, Table 3.34). Increasing B treatments did not affect shoot Na concentration, but significantly decreased shoot content of Na (Table 3.34). Most probably, the decrease in the shoot biomass of medium and high B applied plants resulted in the dilution of the Na in plant tissue [Supanjani and Lee, 2006], [Grieve et al., 2010]. In addition, the significantly lower shoot Na concentration and content of durum wheat compared to bread wheat (Table 3.34) is evidence that mineral nutrient accumulation in plant species can be varied [Alpaslan and Gunes, 2001], [Ismail, 2004]. As mentioned above, salinity tolerance in bread wheat is associated with a locus on the D genome that results in low Na^+ uptake and increased K/Na discrimination.

The negative effect of soil salinity on shoot K concentration (Table 3.29, Table 3.36) and K/Na ratio which are mentioned above in the discussion section of the hydroponic experiments, correlates closely with the soil experiments results [Smith et al., 2010b]. The fact that bread wheat species have a much higher K/Na ratio compared to durum wheat was similarly observed in the two new cultivars added to the second soil experiment (Figure 3.23). The key explanation for this situation that the degree of the reducing impact of salt on the shoot K concentration depends on the plant species [Wang and Wu, 2013], [Läuchli and Grattan, 2007]. As mentioned above, salinity tolerance in bread wheat is associated with a locus on the D genome that results in increased K/Na discrimination. K^+ homeostasis have a critical role in plant salinity tolerance. Mechanism used by bread wheat, namely more superior K^+ retention ability in the leaf mesophyll, also contributes to this distinction [Wu et al., 2014].

Salinity tended to reduce the Ca concentration (Table 3.36). The explanation for the decrease in Ca concentration is the loss of aquaporin activity [Cabañero et al.,

2006] due to the severe increase in Na^+ ions or the decrease in aquaporin expression triggered by salinity stress due to Ca depletion [Bastías et al., 2010]. In addition, the fact that the shoot Ca concentration is higher in durum wheat varieties than in bread wheat cultivars indicates that the Ca concentration may differ between the species [Nuttall et al., 2006], [Del Carmen Rodríguez-Hernández et al., 2013].

Salinity stress has a decreasing effect on shoot Mg concentration (Table 3.36). Although this effect of salinity has been observed in different studies [Grieve et al., 2010], [Ruiz et al., 1997], it has been shown in the literature that the increase in salinity does not always reduce the shoot Mg concentration [Gómez-Bellot et al., 2013], [Bernstein, 1975].

Shoot Mg concentration decreased with increasing B toxicity (Table 3.36). Studies have shown that B toxicity may reduce [Grieve and Poss, 2000] or not affect shoot Mg concentration [Grieve et al., 2010]. Shoot Mg concentration, which is higher in bread wheat species compared to durum wheat species, varies among genotypes [Ruiz et al., 1997], [Escobar et al., 2014], [Sotiropoulos et al., 2006]. Due to the competition between Ca^{2+} and Mg^{2+} ions, bread wheat variety may have higher shoot Ca and lower shoot Mg concentrations than durum wheat variety [Hepler, 2005], [Marschner, 2011].

It is known that the effect of salinity on shoot P concentration may vary depending on factors such as species, cultivar, salt content, salinity level, and media type [Grattan and Grieve, 1998] but no significant effect was observed in this experiment (Table 3.36).

Medium and high salinity reduced the shoot Fe concentration in all three B toxicity treatments and in all four cultivars (Table 3.27). While moderate (15 ppm) B toxicity increased shoot Fe concentration, severe B (30 ppm) application did not affect it (Table 3.37).

While medium salinity (6 $dS.m^{-1}$) increased the shoot Zn concentration, high salt (8.5 $dS.m^{-1}$) application did not affect it (Table 3.37). Furthermore, in literature, main effect of salinity increases Zn concentration in citrus [Ruiz et al., 1997]. A variation in shoot Zn concentration can be observed based on the salinity doses [Grattan and Grieve, 1998]. Soil which contain high pH, low Zn and severe B toxicity have been observed in some semi-arid regions [Cakmak, 2008]. In this thesis, B toxicity application decreased the shoot Zn concentration (Table 3.37). The shoot Zn concentration may differ between plant species and cultivars [Nable et al.,

1997], [Sotiropoulos et al., 2006], [Yau and Ryan, 2008]. Like the hydroponic experiment results, it was seen that the bread wheat variety, Nusrat, had a higher shoot Zn concentration than the other three cultivars (Table 3.37).

Medium and high soil salinity treatment enhanced shoot Mn concentration by similar amounts (Table 3.37). Studies have shown that salinity stress increases shoot Mn concentrations in corn [Gunes et al., 2007a], sugar beet [Khattak and Jarrell, 1989], and rice [Verma and Neue, 1984]. In our study, consistent with the literature and our hydroponic experiments, it was observed that bread wheat cultivars (Nusrat and Adana-99) has higher Mn concentration than durum wheat cultivars (Saricanak and Fuatbey-2000), and therefore it is considered that the shoot Mn concentration may varied between species [Grattan and Grieve, 1998], [Bastías et al., 2010].

As seen in the studies performed, the change in shoot Cu concentration varies depending on the source of salinity and the applied plant species [Rahman et al., 1993], [Izzo et al., 1993], [Grattan and Grieve, 1998]. In the second soil experiment salinity did not affect the shoot Cu concentration (Table 3.37). It was observed that high B toxicity application tended to increase the shoot Cu concentration. Also, there was no consistency determined between cultivars in shoot Cu concentration.

In the second soil experiment performed in this thesis, the application of medium and high soil salinity increased the MDA concentration by similar amounts (Figure 3.24, Table 3.38; Table 3.39). Although no interaction with salinity was noted, main effect of B toxicity increased MDA concentration (Figure 3.24, Table 3.25, Table 3.38). The increase in MDA concentration seen as a result of B toxicity is thought to be due to membrane damage caused by high H_2O_2 level. Similar results have been found in tomato [Cervilla et al., 2007] barley [Karabal et al., 2003], and apple [Molassiotis et al., 2006]. There were minor differences between cultivars and no consistency was noticed among the varieties of bread wheat (Figure 3.26; Table 3.39).

Glycine betaine concentration increased similarly with medium and high salinity application (Figure 3.27, Table 3.39), while proline concentration increased only in plants treated with high salinity (Table 3.39). As salinity induces osmotic stress, an enhancement in the concentration of compatible solutes in plants exposed to salinity stress has been noted in the literature [Agarwal and Pandey, 2004], [Flowers and Colmer, 2008]. In addition, it was determined that proline concentration tended to increase in plants treated with B toxicity (Table 3.39). It is

known that osmoprotectants are not only involved in osmotic adjustment but also in ROS scavenging and membrane stabilization processes [Mansour, 1998], [Munns and Tester, 2008].

It was observed that the total protein concentration increased in plants treated with salinity and B toxicity (Table 3.39). This situation is thought to be related to the concentration effect [Marschner, 2011].

The enzyme activities of SOD, GR, APX, and CAT of Fuatbey-2000, a variety of bread wheat, were found to be significantly higher than other cultivars of wheat (Table 3.40; 3.41). The most important difference of this cultivar compared to others was that the shoot Na concentration was higher in all conditions (Table 3.34). This wheat cultivar, which is exposed to more Na ion stress, therefore be expected to have high antioxidative enzyme activities as a stress response.

While the activities of SOD and APX enzymes increased, GR activity decreased in B-toxic plants (Table 3.41). Also, CAT activity increased and decreased depending on the dose of B toxicity. The oxidative effect of B toxicity was determined in this soil experiment conducted in greenhouse. The reasons for this situation can be considered as semi-controlled greenhouse conditions, which are the growth environment of the plants, and high light intensity which increases the stress severity [Mittler, 2002], [Cakmak and Marschner, 1992]. It has been observed in the literature that B toxicity increases SOD, APX, and CAT enzyme activities depending on the plant species and dose [Cervilla et al., 2007], [Ardic et al., 2009]. Since the H_2O_2, the product of SOD enzyme activity, is the substrate of APX and CAT enzymes, a correlation was expected between each other. Increased salinity did not affect SOD and GR activities, it decreased APX and increased CAT enzyme activities (Table 3.41).

Specific SOD and APX activities decreased with increasing salinity stress (Table 3.42). The sp. activities of these two enzymes, which have a product-substrate relationship with each other, may have decreased as a result of increasing total protein concentration due to salinity [Molassiotis et al., 2006]. In addition, it is known that total protein concentration increases with B toxicity (Table 3.39). The fact that sp. CAT and GR activities decrease as the B toxicity increases, can also be explained by increased total protein concentration [Mittler, 2002]. However, the sp. SOD activity varies depending on the severity of B toxicity, and decreased in plants

applied medium B, but increased in plants treated with high B [Hamurcu et al., 2016], [Cervilla et al., 2007].

In the third soil experiment, which has a similar plan with the second soil experiment, the REL was evaluated before the yield parameters were examined. It has been determined that both salinity and B toxicity increase REL separately (Figure 3.29), thus membrane integrity is disrupted and membrane permeability is excessively increased [Zhu et al., 2004], [Karabal et al., 2003]. We have seen above that B toxicity increases MDA concentration and some antioxidant enzyme activities. One of the reasons for these rises may be that, by disrupting living membranes, toxic B raises REL. The amount of REL, which can vary greatly between wheat varieties and even cultivars [Tavallali et al., 2017], is listed in this experiment as follows: Saricanak, Adana-99, Nusrat, and Fuatbey-2000 (Figure 3.29). In the amount of REL among durum wheat varieties, there appears to be no consistency, and the very low REL amount of Fuatbey-2000 cultivar can be associated with the high enzyme activities we saw as a result of the previous experiment.

Chlorotic and necrotic symptoms observed at the tips and edges of the leaves before senescence in 90-days-old plants were scored and their results were recorded (Table 3.43). It was expected that the symptom score would increase as a result of toxic B treatment [Nable et al., 1997]. Despite the decline in straw B concentration with increasing salinity (Table 3.45), why was there no improvement in symptom score? This may be since plants grow in a semi-controlled environment with high light intensity and face undefined stress factors. Probably for these reasons, although the salinity decreased the B concentration, it did not affect the symptom score.

In previous experiments, mineral nutrients were evaluated only in the shoot and root of plants in the early vegetative period, while in the third soil experiment mineral nutrients in mature plants were measured in straw and grain after senescence.

Just like the shoot (Table 3.33), the straw B concentration (Table 3.45) was elevated in plants treated with B toxicity while it decreased with increasing salinity. The antagonistic relationship between salinity and B toxicity was observed in straw. However, when the grain B concentration was evaluated, it was observed that B toxicity increased the B concentration in grain, but there was no antagonistic relationship with salinity (Table 3.49).

The following question emerges from above situation: Why did the interaction of salinity and B toxicity not have the same kind of effect on straw and grain on B

concentration? To answer this question, we must first know that the vascular bundles that transfer B to shoot (source) and grain (sink) tissues are different [Nable et al., 1997]. While most of the B taken from the media is carried to the leaves by xylem, the B in the leaves is transported to the grain by phloem [Raven, 1980]. These results lead us to suggest those,

1) Restriction of remobilization of B transported from source to sink by phloem.
2) Phloem mobility of B depends on the species and is very low in wheat.
3) Limited sink activity regardless of the mineral nutrient.

It is very usual to see a difference in the mineral concentration of the vegetative and generative tissues because the factors contributing to B accumulation in straw and grain are different [Brown and Shelp, 1997]. As a result of transpiration restricted by rising salinity, root to the shoot B transport by xylem decreased, while B transport shoot to grain by phloem was not affected by salinity.

Shoot (Table 3.33) and straw (Table 3.47) B concentration was higher in durum wheat than bread wheat. Depending on the species, the accumulation of B in above-ground tissues may differ [Nable et al., 1997]. This disparity between bread and durum wheat species could be due to the difference in the transpiration of the plants. As can be seen in the hydroponic experiment results, the transpiration of Saricanak is higher than Nusrat (Table 3.21, Figure 3.13). Higher accumulation of shoot boron in durum wheat compared to bread wheat is a known phenomenon [Kalayci, 1998].

As salinity increases, the rising concentration of Na in straw (Table 3.45) is expected to enhance the concentration of grain Na (Table 3.49). Since Na is not a phloem-immobile nutrient such as B, transport of Na occurs linearly from shoot to grain by phloem, depending on the sink activity [Francois et al., 1986], [Holloway and Alston, 1992]. Straw and grain Na concentration, which is higher in durum wheat varieties, may be related to transpiration, just like B (Table 3.21; Table 3.45). Also, durum wheat is much more salt-sensitive than bread wheat, owing to its low Na^+ exclusion capacity [Munns, 2006].

Straw Na concentration tended to decrease slightly as a result of B toxicity application to plants under saline conditions (Table 3.45). However, a similar antagonism was not seen in the grain Na concentration results (Table 3.49). The most important factor affecting this situation is the vascular bundle type that carries the

related nutrient [Läuchli and Grattan, 2007]. Since the transport of Na to the sink tissue is through phloem and despite Na is not a phloem-immobile, there is no such antagonism in grains due to low sink activity.

In the plant grown in saline-soil, K uptake decreases due to the competition between Na^+ and K^+ [Sauer et al., 2013]. Therefore, a reduction in straw K concentration is expected in plants grown in saline soil (Table 3.45). In addition, increasing salinity typically reduces the K/Na ratio of straw as the salinity declines the K concentration and increases the Na concentration (Figure 3.30). In described before, K^+ homeostasis have a critical role in plant salinity tolerance. Mechanism used by bread wheat, namely more superior K^+ retention ability in the leaf mesophyll, also contributes to this distinction [Wu et al., 2014]. In the previous paragraph, it was discussed that the concentration of straw Na tends to decrease in B-toxic plants (Table 3.45). On the contrary, straw K concentration tended to increase in plants treated with B toxicity (Table 3.45). It is not surprising that increased K concentration in straw, where Na was reduced due to Na mimicking K [Grattan and Grieve, 1999]. The fact that increasing B toxicity tends to improve the K/Na ratio can also be explained by this scenario (Figure 3.30).

We know from previous results that durum wheat straw Na concentration is higher than bread wheat (Table 3.45). The higher straw K concentration in bread wheat species can also be explained by the antagonistic relationship between K-Na [Grieve and Poss, 2000]. This antagonistic relationship was of course reflected in the results of the K/Na ratio and it was observed that bread wheat varieties had a higher K/Na ratio (Figure 3.30). The opposite situation was observed in the grain K concentration and it was determined that the grain K concentration of durum wheat cultivars was higher than that of bread wheat species (Table 3.49).

As seen in the literature [Francois et al., 1986], increasing salinity gradually decreased the straw dry weight in all cultivars (Table 3.46; Table 3.47). While medium B toxicity did not have a significant impact, the straw dry matter in high B treated plants was significantly reduced and a synergistic relationship between salinity and B toxicity was observed (Table 3.47). In the present study, the decrease in wheat growth with increasing salinity and B levels were most probably due to the disruption of various metabolic processes such as photosynthesis, transpiration and stomatal conductance, subsequently resulting in poor growth leading to low biomass [Naz et al., 2019], [Manchanda and Sharma, 1991].

It was determined that the grain yield is significantly reduced by salinity and high B applications and there is no interaction between them in this respect (Table 3.46, Table 3.47). As a consequence of the negatively affected parameters by stress agents during the vegetative period, primarily the photosynthetic area, reproductive development was impaired [Grieve and Poss, 2000]. Some studies suggest that the grain yield is reduced by salinity and B toxicity in an additive and antagonistic manner, respectively [Manchanda and Sharma, 1991], [Holloway and Alston, 1992].

Harvest index (HI) is briefly known as the efficiency of grain production [Gholizadeh et al., 2014] and in this thesis among the stress agents and different doses, only high salinity tend to reduce the HI (Table 3.46). The fact that B toxicity does not adversely affect or even tend to increase the HI (Table 3.47) is attributed to the reason that vegetative growth is more sensitive to B toxicity than generative growth [Princi et al., 2015].

It was observed that both grain yield and HI of bread wheat species were significantly higher than durum wheat species (Table 3.47). The genotypic variation lies at the basis of this difference between bread and durum wheat species [Cossani et al., 2011]. Bread wheat generates more grain yield than durum wheat, as expected for evolutionary and breeding reasons, and this appears to be a constitutive distinction between species [Marti and Slafer, 2014]. The variations in graining efficiency, the number of grains produced per unit of spike dry weight at anthesis, could form the basis for the constitutive differences in grain yield and HI.

High salinity and high B toxicity significantly reduced the number of spikes (Table 3.48). Spike formation interrupted by stress agents will cause low yield [Gholizadeh et al., 2014] and similar results have been observed in the literature [Grieve and Poss, 2000], [Maas and Grieve, 1990]. Although Saricanak spike number is significantly higher than other cultivars, the difficulties in grain forming and grain filling are the reason for having lower grain yield compared to bread wheat species.

Soil salinity decreased grain number gradually but did not affect thousand grain weight (TGW) (Table 3.48). Therefore, we can realize that while salinity inhibits grain formation, it did not significantly affect grain filling [Cossani et al., 2011]. Severe B toxicity, like salinity, diminished the grains developed in the spike and thus decreased the number of grains (Table 3.48). It was also noticed that B toxicity did not reduce TGW, so it did not have a negative impact on grain filling,

and even moderate B toxicity treatment tended to increase TGW. Therefore, it was thought that there might be B deficiency in the control soil used in the experiment.

When compared, it was observed that bread wheat species had significantly higher grain number and TGW than durum wheat species (Table 3.48). Therefore, it was seen that bread wheat was more efficient in terms of grain set and grain filling, which are the most important yield parameters. As anticipated for evolutionary and breeding factors, bread wheat generates more grain number and TGW than durum wheat, and this appears to be a constitutive distinction between species [Marti and Slafer, 2014], [Cossani et al., 2011]. Grain formation efficiency differences and the number of grains produced per unit of dry spike weight at anthesis, could form the basis for the constitutive differences in the number of grains and TGW.

In general, shoot and root growth of both bread and durum wheat species were not affected by salinity and B toxicity combination under hydroponic and soil cultures. Salinity and B toxicity interaction additively affect these plant growth parameters. Increased B toxicity enhanced B accumulation and necrosis severity in both bread and durum wheat shoot tissues but decreased with increased salinity under both hydroponic and soil culture. Also, increased severity of B toxicity tended to reduce shoot Na accumulation. Antagonistic (alleviating) relationship was observed between salinity and B toxicity on toxic ion accumulations and necrosis severity in bread and durum wheat species under hydroponic and soil culture conditions. It was observed that durum wheat cultivars accumulate more toxic ions and more sensitive to salinity and B toxicity than bread wheat cultivars. Salinity and B toxicity combination has no significant effect on oxidative and osmotic stress parameters under hydroponic and soil cultue conditions. Therefore, salinity and B toxicity combination additively affect these stress parameters. Under greenhouse conditions, photooxidative stress occurred and aggravated negative effects of stress parameters. Yield parameters were additively affected by salinity and B toxicity combination. In addition, grain B and Na accumulation was not affected antagonistically by salinity and B toxicity like shoot and straw. Also, durum wheat cultivars have lower yield parameters than bread wheat. Based on these informations, this thesis study explained the inconsistencies in the literature.

5. CONCLUSION

Both salinity and boron (B) toxicity are among major abiotic stress factors that adversely affect crop production and thus pose a threat to the economy as well as global food security. Often, they co-occur in fields in arid and semi-arid regions, especially where agriculture depends on irrigation with poor quality water with a high salt and B content. Due to complex interactions between them, the effects of the combination of salinity and B toxicity on crops may be different from the individual effects of these factors. Although this interaction has been subject to a lot of studies in the literature, there are inconsistencies in results and the physiological factors underlying them are largely unknown.

For this purpose, bread (*T. aestivum* cv. Nusrat and Adana-99) and durum (*T. durum* cv. Saricanak and Fuatbey-2000) wheat species were grown with different salinity and B supply levels in soil and hydroponic culture under a controlled growth chamber and greenhouse conditions. The effects of stresses alone and in combination on vegetative growth and the necrosis in the leaves were determined. Data on the B and Na accumulation and distribution in the young and old leaves and root tissues were collected by ICP-OES. Transpiration was measured and the correlation between transpiration and toxic element accumulation was investigated. The damage caused by stress factors to membrane integrity was determined by relative electrolyte leakage analysis. Lipid peroxidation analysis and total and specific activity measurements of catalase, ascorbate peroxidase, glutathione reductase, and superoxide dismutase enzymes analysis were performed to determine oxidative damage caused by stresses separately and together. Also, concentrations of sugar alcohols, proline and glycine betaine was determined.

Salinity was affected shoot growth and B toxicity was affect root growth and salinity-B toxicity interaction have additive effect on shoot and root growth. It could be explained by necrotic lesions in source tissues declines photosynthetic area and so decrease translocation of photoassimilates to sink tissues like roots. Although toxic B affects root growth, it was found that it did not accumulate in the root but accumulate at the tips of the leaves and this accumulation and related necrosis and membrane damage decreased with salinity. This stuation could be explained by reducing effect of salinity on transpiration. Similar B concentrations were found in the shoot of the

selected cultivars while higher Na concentrations were measured in the tissues of durum cultivars. With or without salinity, B toxicity did not contribute to the osmoprtectant accumulation and oxidative injury of the plant shoots under growth chamber conditions. However, under greenhouse conditions, osmoprotectant accumulation, lipid peroxidation, and antioxidant enzyme activities were increased by B toxicity. The difference here is related with growth conditions. Photooxidative stress increased due to the plants grown in the greenhouse because of the plants exposed to high light intensity. Also, it was observed that the effect of salinity and B on vegetative growth is more severe than generative development. Boron toxicity treatments tended to reduce yield parameters, especially the grain yield and harvest index, also salinity stress reduced these parameters significantly. Salinity and B toxicity combination have additive effect on yield parameters. The reason behind the reduction of yield by salinity and B toxicity is decreased grain numbers in stress-applied plants. Although increased grain size, eventually, yield declined because of the reduction of grain numbers. Moreover, the yield parameters of durum wheat cultivars were detected quite lower than that of bread wheat. Based on these informations, this thesis study explained the inconsistencies in the literature.

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