THE INFLUENCE OF SOIL SALINITY GRADIENTS ON SOYBEAN [*GLYCINE MAX* (L.) MERR.] AND CORN (*ZEA MAYS* L.) GROWTH

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The Influence Of Soil Salinity Gradients On Soybean [*Glycine Max* (L.) Merr.] And Corn (*Zea Mays* L.) Growth

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ABSTRACT

An estimated 2.3 million hectares are salt-affected in North Dakota (Brennan and Ulmer, 2010), a number increasing due to land management, climate, and crop choice. As a result, yield reductions are noted for salt-sensitive crops such as soybean [*Glycine max* (L.) Merr] and corn (*Zea Mays* L.). The objective of this greenhouse study was to assess soybean and corn response to salinity, using sulfate based salts. Soybean leaf area, plant mass, and height decreased by 66, 59, and 47%, respectively, across a salinity gradient ranging from an $EC_{1:1}$ of 0.4 to 4.1 dS m^{-1}. Corn mass and height decreased by 42 and 26%, respectively, root length and mass also decreased by 44 and 37%, respectively from an $EC_{1:1}$ 0.8 to 5.3 dS m^{-1}. Thus, planting soybean and corn on salt-affected soils in North Dakota will result in overall decreased productivity for both crops even at low levels of salinity.

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GENERAL INTRODUCTION

Soil salinity is a global problem impacting land productivity and food security. Land degradation due to salinity is common in arid and semi-arid regions where evaporative demand exceeds potential precipitation, leading to an accumulation of soluble salts in to the root zone (Ghassemi et al., 1995). Additionally, 20% of irrigated lands are salt-affected (Ghassemi et al., 1995). Shifts in climate and agronomic practices have also increased the amount of salt-affected soils with time (Metternicht and Zinck, 2003). This worldwide trend of salinization (through a variety of processes) extends to the northern Great Plains region of the United States. Here, cropping practices have shifted to favor non-salt tolerant crops such as soybean [*Glycine max* (L.)] and corn (*Zea Mays* L.) and this has made soil salinization a primary concern.

Increased precipitation in North Dakota since 1993 has resulted in higher water tables, redistribution of salts, and other production issues (Scherer and Jia, 2010). Though drought conditions plagued the state from 1988 to 1992 (Ashworth, 1999), current increased precipitation since the early 1990's has shifted water resources, by bringing the depth to water closer to the surface and increasing water transport across the landscape. Beyond landscape scale salt movement, soluble salts are able to move vertically in the soil (Franzen, 2013). Capillary rise, due to evaporative demand allows for the upward movement of soil water to the surface (Smiles and Philip, 1978; Hillel 1998; Isidoro and Grattan, 2010), which can contribute to increased salinization in the root zone.

As soil salinity has increased in the region, farming practices have also evolved. Larger equipment, shifts in crop prices, and changing crop genetics combined, has led to management and cultural changes in agriculture. Regionally, crop rotations have shifted from salt-tolerant small grain crops to include less tolerant species, i.e. soybean (*Glycine Max* (L.) Merr.) and corn (*Zea Mays* L.) (National Agricultural Statistics Service, 2014).

Soybean has become an increasingly important crop in North Dakota. The 2014 Survey of Agriculture reported 2.4 million hectares of soybean harvested in North Dakota having $1.9 billion in production value (National Agricultural Statistics Service, 2014). Comparatively, 1997 Census data reported only 462,920 hectares of soybean harvested (National Agricultural Statistics Service, 2014). Corn has also become a valuable commodity crop in North Dakota. The 2014 Survey of Agriculture reported approximately 1 million hectares of corn harvested in North Dakota, worth an estimated $1 billion in production value (National Agricultural Statistics Service, 2014). Whereas in, 1997 Census data reported 200 thousand hectares of corn were harvested (National Agricultural Statistics Service, 2014), a difference of over 800 thousand hectares of corn in the state of North Dakota in less than twenty years.

Current estimates are that roughly 2.3 million hectares of land in North Dakota are salt-affected, a number which has increased over the past two decades(Brennan and Ulmer, 2010). Salinization of soils in this northern region is closely linked to a combination of the (1) abundance of naturally occurring soluble salts, (2) a shift in the hydrologic cycle and (3) agricultural management practices (Anderson et al., 2010). The combination of local farming practices, i.e. growing crops not suited for saline soils, and underlying geologic makeup are leading to increasing salinity levels and decreased land productivity. The purpose of this thesis is to assess soybean and corn response to salinity using greenhouse methodology designed to resemble North Dakota salinization, i.e. soluble salts accumulating in the root zone via capillary rise and landscape movement, to better determine corn and soybean tolerance to saline soils of North Dakota.

JUSTIFICATION

The United States Department of Agriculture (USDA) Agriculture Research Service's (ARS) Salinity Laboratory released an assessment of crop salt tolerance in 1977 (Maas and Hoffman, 1977). In the report salinity tolerance was reported for many crops, including soybean and corn. Threshold levels, where a decline in yield was observed, were 5.0 and 1.8 dS m^{-1}, electrical conductivity from the saturated paste (EC_e), for soybean and corn, respectively. Threshold levels stated by the U.S. Salinity Lab were assessed using NaCl based salts for soybean (Abel and Mackenzie, 1964; Bernstein and Ogata, 1966) and NaCl and $CaCl_2$ salts for corn (Kaddah and Ghowail, 1963). Numerous studies since the early 1950's and 60's have focused on plant response to salinity; however, most methods use chloride based salts. Furthermore, most studies involve application of salt treatment through irrigation. Salinity in North Dakota is caused primarily by sulfate based salts and the capillary rise of salts into the rooting zone; therefore, prior research using chloride salts and irrigation methods may not be an accurate assessment of corn and soybean response to salinity in North Dakota.

LITERATURE REVIEW

Plant Response to Salinity

The productivity of plants is largely influenced by abiotic stress. Projections estimate that 50% of plant yield reductions are a result of abiotic stressors, including salinity, drought, temperature, flooding, etc (Rodriguez et al., 2005). Among abiotic stressors, soil salinity is one of the more destructive factors limiting the productivity of crop plants (Hasanuzzaman et al., 2013). Most crop plants are sensitive to high levels of salt concentrations in the soil, due to physiological effects. Numerous functional groups are affected by salt stress in plants, including: proteins, ion transport, energy metabolism, carbohydrates, cytoskeleton, lipid metabolism, and stress related hormones, among others (Kosova et al., 2013).

In regards to salt tolerance, there are two categories of plants: halophytes and glycophytes. The Greek root word *halo-* means “from salt”, therefore, halophytes are plants that can survive, and even thrive, in saline soils. These specialized plants account for only 2% of all terrestrial species, most of which belong to the higher plant families (Glenn and Brown, 1999). Plants without the adaptations required to survive in saline environments are termed glycophytes, from the Greek root word *glycol-* meaning sugar. Curiously, glycophytes and halophytes seem to have evolved similar mechanisms to deal with salts, halophytes have simply developed more efficient mechanisms than their counterparts (Kosova et al., 2013). Furthermore, halophytes are a very diverse group and are found in many plant families. Their unique adaptations are thought to have arisen during the diversification of angiosperms; thus, halophytes are not limited to a specific taxon or region (O’Leary and Glenn, 1994).

Physiological and morphological salinity tolerance mechanisms shared by halophytes and glycophytes include: osmotic adjustment, ion exclusion, compartmentalization, and excretion.

Osmotic adjustment is the primary mechanism salt tolerant plants use to cope with salt stress, e.g. accumulation of Na^+ in the leaf tissue to facilitate the osmotic adjustment (Glenn and Brown, 1999). Some species are able to exclude virtually all salts from entering the plant, with thickened layers of suberin, a waxy coating, on the epidermal cells of the root (Glenn and Brown, 1999). In other cases, salts move into the plant with water uptake, but are compartmentalized into the vacuoles of the cells. Compartmentalization prevents interference with any cellular function (Kosova et al., 2013). Furthermore, halophytes actively scavenge for Na^+ to sequester into the vacuole, and while glycophytes are able to sequester salts into vacuoles, the process is not as efficient (Glenn and Brown, 1999). Excretion of salt is a secondary salt tolerance mechanism, plants excrete salts through salt glands, salt bladders, and succulent tissues. The halophytes are a complex group, even within the same family and their variation in adaptions to salt tolerance is vast. For example, black mangroves (*Avicennia germinans*) tolerate salt, in part, due to excretion through salt glands. In contrast, red mangroves (*Rhizophora mangle*), equally as tolerant to salts, do not excrete salts but rely on other mechanisms of salt tolerance (Glenn and Brown, 1999).

As salt-affected land increases worldwide (Ghassemi et al., 1995), interest in improving the salt tolerance of crops is increasing. However, this is not a simple task since salt tolerance adaptations are complex in halophytes, ranging from cellular level systems to the plant function as a whole. Therefore, inserting a single trait of a halophyte into a glycophyte is not possible at this time (Flowers and Yeo, 1995). Direct domestication of halophytes may be the more immediate way to move forward with crop production on saline, degraded land. Halophytes have shown great promise as forage and oilseed crops (Glenn and Brown, 1999) and although halophytes have shown marketable potential as crops, adoption of these species is limited.

Crop Response to Salinity

The vast majority of the world's agricultural crops are glycophytes, and are not suited for production on saline ground. Furthermore, very few patents have been issued for varieties listed as salt tolerant (Flowers and Yeo, 1995). Day (1987) developed a corn variety said to be suited for saline environments. Three chrysanthemum, two alfalfa, a meadow cord grass, and three salt grass varieties were patented as salt tolerant (Dobrenz et al., 1989; Johnson et al., 1991; Flowers and Yeo, 1995). The lack of germplasm developed for salinity tolerance can be attributed to the complexity of salt tolerance mechanisms (Kosova et al., 2013) and, potentially, the lack of interest and resources among plant breeders (Flowers and Yeo, 1995).

Crops range in salt tolerance and thus, symptoms of salt stress vary among crops (Maas and Hoffman, 1977). In general, salt-stressed crops are stunted, occasionally leaves develop a darker green hue and succulence, and woody species can develop necrosis on leaves, however, this has not be noted in herbaceous crops (Maas and Hoffman, 1977). Crops also vary in their tolerance based on stage of growth: e.g. barley, wheat, and corn are most sensitive at germination and early growth stages (Kaddah and Ghowail, 1964), whereas rice is most tolerant at germination in contrast to later growth stages (Kaddah, 1963). Climate plays a role in crop salt tolerance as well. Hot and dry conditions tend to reduce a crop tolerance, compared to cool and humid climates that can increase relative salinity tolerance (Hoffman and Rawlins, 1970).

Crop response to salinity can be broken into two phases (1) osmotic stress and (2) ion specific injury (Munns, 2002). There is much debate concerning osmotic stress versus ion-specific stress (Bernstein, 1975). The osmotic "school of thought" attributes the adverse effects of salinity to be primarily an effect of decreased osmotic potential, which is caused by soluble salts in the soil solution. Conversely, the ion-specific "school of thought" considers stress imposed by individual ions to be the main explanation. Based on past research, it is clear that both osmotic and ionic stresses are involved in overall crop response to salinity. As salt concentration increases in the soil solution, the osmotic potential of the soil decreases while osmotic pressure of the plant increases which limits water uptake and induces a drought-like stress. Plant response to osmotic stress occurs within minutes to hours of salinization where leaves exhibit an immediate reduction in growth due to changes in cell water relations (Munns, 2002). Neumann (1993) found this immediate reaction in corn when exposed to NaCl. After a plant has adjusted to the osmotic stress, ion specific injury can occur (Bernstein, 1975).

Ion-specific toxicity can occur at high salinity levels at which time salt sensitive plants can no longer exclude salts from entering through the root tissue and the salt concentration within the plant increases, leading to ion-specific injury on the leaves within days (Munns, 2002). Typically, injury is found on the older transpiring leaves, similar to symptoms for a mobile nutrient deficiency. Like halophytes, glycophyte crops have the mechanisms to exclude salts from entering at the root and compartmentalize in vacuoles, however these mechanisms are often termed 'leaky' (Hasanuzzaman et al., 2013) and allow salts to enter the plant resulting in eventual toxicity. For example, the walls of vacuoles in halophytes are often thickened to prevent leakage of salts stored there (Flowers and Yeo, 1945). Most crops do not have this adaptation and compartmentalized salts can leak into the cytoplasm and alter cell function (Munns, 2002). Ultimately, glycophyte production is predominately limited by osmotic stress, along with high ion uptake and limited compartmentalization.

Salinity Tolerance Testing

Although most crops are categorized as glycophytes, there is a range of tolerance among crops. Certain crops are better suited for production on salt affected soils than others. The USDA ARS Salinity Laboratory in Riverside, CA conducted and compiled many of the crop tolerance curves currently used (Maas and Hoffman, 1977). Canada's salt tolerance testing laboratory in Saskatchewan also produced and refined the crop tolerance knowledge base, using a sophisticated controlled testing facility (Steppuhn and Wall, 1999). Additional research on crop tolerance and response to salinity as been published in numerous scientific journals, sourced from across the world and thus, the knowledge base is significant.

Many of the crop tolerance levels established in 1977 by the USDA-ARS Salinity Laboratory in the 1970's are the standard today. The 1977 report was a compilation of literature and data from 1950 to 1975 (Maas and Hoffman, 1977). Mass and Hoffman (1977) proposed a curve composed of two segments (1) a tolerance level with a slope of zero and (2) a line with a slope representing yield reduction per unit of salinity. The crop tolerance line represents the maximum amount of salinity the crop can sustain without yield reduction. Beyond the tolerance level relative yield declines along the slope, as salinity increases. Based on the tolerance level and slope, relative yield of a crop can be determined at any given salinity level (measured as the electrical conductivity of the saturated paste extract, EC_e) using Equation [1], where B is the slope, EC_e is the salinity, A is the crop threshold in dS m^{-1}, and Y is the predicted relative yield (Maas and Hoffman, 1977).

$$Y = 100 - B\ (EC_e - A) \qquad (1)$$

Equation (1) is useful for determining crop response at a specific salinity level. The report also categorizes crops in the following categories according to their tolerance levels: sensitive ($EC_{1:1}$ = 0.0 – 4.4 dS m^{-1}), moderately sensitive ($EC_{1:1}$ = 4.5 – 8.9 dS m^{-1}), moderately tolerant ($EC_{1:1}$ = 9.0 – 13 dS m^{-1}), and tolerant ($EC_{1:1}$ = 13 – 18 dS m^{-1}). Maas and Hoffman (1977) report salinity tolerance as the electrical conductivity from the saturated paste (EC_e). For consistency values have been converted from EC_e to the EC from a 1:1 soil to water ratio ($EC_{1:1}$) for this thesis using the conversion described by Smith and Doran (1996).

Canada's salinity testing lab was constructed with the goal of determining salinity tolerance of regional crops and varieties for dryland and irrigated agriculture (Steppuhn and Wall, 1999) and is one of only two laboratories of its kind in North America. The laboratory sowed seeds directly into saline seedbeds to replicate dryland scenarios (Steppuhn, 2013), in contrast to the more common method of irrigating with salt water to achieve treatment levels. Canada's salt laboratory redefined salt tolerance for the crops of the region, namely: wheat (*Triticum aestivum* L.), pea (*Pisum sativum* L.), canola (*Brassica napus*), and camelina (*Camelina sativa*). Table 1 summarizes methods and tolerance results from an array of studies on soybean, corn, and other closely related crops.

As illustrated in Table 1, the majority of studies are conducted in a greenhouse, using chloride-based salts. Variation in testing methods and interpretation for tolerance levels can have significant implications on agriculture and crop choice. For example, Maas (1993) reported wheat threshold level at an $EC_{1:1}$ of 3.4 dS m^{-1} using irrigation greenhouse methods, whereas Steppuhn and Wall (1999) determined the threshold levels for two wheat varieties to be 0.8 to 1.4 dS m^{-1} ($EC_{1:1}$) using dryland testing methods. Thus, the method used to estimate a threshold level should be factored into an interpretation of that value.

Table 1. Compiled methodologies and results from previous soybean [*Glycine max* (L.) Merr.], corn (*Zea Mays* L.), and related crop salinity response greenhouse studies.

Crop	Salt Type	Method	$EC_{1:1}$ Range†	% Decline from low to high salinity treatment‡	Reference
			dS m^{-1}		
Soybean	NaCl	Greenhouse, irrigation	0.0 - 5.0	78% yield	Ghassemi-Golezani and Taifeh-Noori, 2011
Soybean	NaCl	Greenhouse	0.0 - 79.9 mM	- §	Singleton and Bohlool, 1984
Soybean	NaCl	Greenhouse	1.7 - 7.7	-	Abel and Mackenzie, 1964
Soybean	NaCl; $CaCl_2$	Greenhouse, irrigation	0.4 - 3.9	39% biomass; 46% yield	van Hoorn et al., 2001
Soybean	NaCl	Greenhouse, irrigation	0.0 - 1.6	-	Tu, 1981
Soybean	NaCl; $CaCl_2$	Greenhouse, irrigation	0.8 - 5.4	21% biomass; 29% plant height	Shalhevet et al., 1995
Soybean	NaCl	Greenhouse, irrigation	0.4 - 4.3	75% plant height	Bustingorri and Lavado, 2011
Soybean	NaCl	Greenhouse	0.0 - 5.4 atm	-	Bernstein and Ogata, 1966
Bean, Broad	NaCl; $CaCl_2$	Greenhouse, irrigation	0.4 - 3.7	18% biomass; 50% yield	van Hoorn et al., 2001
Bean, Common	NaCl; $CaCl_2$	Greenhouse	1.3 - 2.9	-	Adiku et al., 2010
Bean, Pinto	NaCl; $CaCl_2$	Greenhouse, dryland	0.7 - 13.9	100% biomass and height	Steppuhn et al., 2001
Lentil	NaCl; $CaCl_2$	Greenhouse, irrigation	0.4 - 1.7	73% biomass; 88% yield	van Hoorn et al., 2001
Pea	NaCl	Greenhouse	0.0 - 5.6	10 to 45% biomass ¶	Shahid et al., 2012
Pea, Green	NaCl; $CaCl_2$	Greenhouse, dryland	0.7 - 13.9	100% biomass; 99% plant height	Steppuhn et al., 2001
Pea, Yellow	NaCl; $CaCl_2$	Greenhouse, dryland	0.7 - 13.9	100% biomass and plant height	Steppuhn et al., 2001
Corn	NaCl	Greenhouse, irrigation	0.0 - 1.5	22% biomass; 8% plant height	Bilgin et al., 2008
Corn	NaCl; $CaCl_2$	Greenhouse, irrigation	1.0 - 5.3	14% biomass; 36% plant height	Shalhevet et al., 1995
Corn	Na_2SO_4; $MgSO_4$;$CaCl_2$	Greenhouse, irrigation	0.0 - 8.7	-	Hassan et al., 1970
Corn	NaCl; $CaCl_2$	Field, irrigation	0.5 - 2.0	21% yield	Katerji et al., 2004
Corn	NaCl; $CaCl_2$	Field, irrigation	0.7 - 5.0	-	Shani and Dudley, 2001
Corn	NaCl; $CaCl_2$	Greenhouse, irrigation	0.2 - 5.0	-	Khalil et al., 1967
Corn, Sweet	NaCl; $CaCl_2$	Field, irrigation	0.3 - 4.2	-	Shenker et al., 2003
Corn	NaCl; $CaCl_2$	Field, irrigation	0.0 - 7.9	48% yield	Kaddah and Ghowail, 1963
Sorghum	NaCl	Greenhouse, irrigation	0.0 - 8.4	62% biomass	Netondo et al., 2004
Sorghum	NaCl; $CaCl_2$	Field, irrigation	1.7 - 6.9	92% yield; 31% biomass; 60% plant height	Francois et al., 1984

† $EC_{1:1}$ is estimated based on a medium soil texture from Smith and Doran (1996)

‡ Estimated based on data reported in cited reference

§ Data not reported in reference

¶ Range among pea genotypes shoot dry wt. in response to salinity

Salt type also varies among studies. Chloride-based salts are commonly used, with NaCl and $CaCl_2$ being the typical mix, while sulfate (SO_4) based salts are rarely used (Table 1). Sulfate and Cl-based salts can have similar effects on growth and osmotic restrictions, however, differences exist in physiological response of plants to the specific ion (Maas and Nieman, 1978). Maas and Nieman (1978) report that Cl-based salts cause plant epidermal cells to enlarge, leaf surface area to decline, and limited cell division. In contrast, SO_4 salinity can inhibit cell expansion, cause decreased succulence, and increase stomata numbers, while maintaining cellular division rates (Strogonov, 1962). Abel and MacKenzie (1964) attributed varietal differences in soybean tolerance to the diversity of Cl transport mechanisms among varieties. Since Cl transport largely influences Cl based salinity tolerance, SO_4 salinity tolerance may differ drastically. Furthermore, Cl is readily absorbed by the plant and contributes more to osmotic adjustment than SO_4 (Maas and Nieman, 1978). Overall, Cl and SO_4 salts affect plants differently.

Experimental methods greatly influence the potential interpretations of salinity tolerance results. The consistent use of tolerance interpretation based on Cl salts could be inaccurate in regions largely dominated with non-Cl based salts, e.g. Northern Great Plains. Regardless of how a soil becomes salinized the ionic composition of salinity can vary by region. In North Dakota, cretaceous shales/sandstone and Ordovician limestone are the source of SO_4-based soluble salts (Benz et al., 1976). Sulfate salts are the dominant salt form across the upper Great Plaines; with Cl salts found in the northeast section of the state (Benz et al., 1976; Franzen, 2013). For the majority of North Dakota, salinity threshold levels should be interpreted with the consideration that salt type differs.

Production Implications of Saline Soils in North Dakota

North Dakota has an estimated 2.3 million hectares of saline soil (Brennan and Ulmer, 2010). Crops in grown in North Dakota range from salt tolerant, e.g. barley (*Hordeum vulgare* L.) to sensitive, e.g. soybean (Table 2). Wheat ranks highest in harvested hectares across the state (National Agricultural Statistics Service, 2014), and as a moderately tolerant to tolerant crop, wheat can be productive on these salt-affected soils. In recent years, markets have pushed the production of more salt sensitive crops, e.g. soybean and corn compared to more salt tolerant crops, e.g. wheat and barley (National Agricultural Statistics Service, 2014). Productivity will be limited for these salt sensitive crops on the saline soils of North Dakota. The objective of this thesis is to assess soybean and corn response to salinity for North Dakota's salt-affected hectares so that farmers and landowners have increased knowledge on selecting crops.

Table 2. Salinity tolerance for the major crops grown in North Dakota.

Crop	Scientific Name	Threshold EC_e	Threshold $EC_{1:1}$†	Slope	Tolerance Rating‡	Reference
		dS m^{-1}		% per dS m^{-1}		
Barley	*Hordeum vulgare* L.	8.0	4.5	5.0	T	Ayars et al., 1952; Hassan et al., 1970
Canola	*B. napus* L.	11.0	6.2	13.0	T	Francois, 1994
Corn	*Zea mays* L.	1.7	1.0	12.0	MS	Bernstein and Ayars, 1949; Kaddah and Ghowail, 1964
Dry Beans	-	1.0	0.6	19.0	S	Osawa, 1965
Flax	*Linum usitatissimum* L.	1.7	1.0	12.0	MS	Hayward and Spurr, 1944
Potatoes	*Solanum tuberosum* L.	1.5	0.8	14.0	MS	Bernstein et al., 1951
Soybean	*Glycine max* (L.) Merr.	5.0	2.8	20.0	MT	Abel and McKenzie, 1964; Bernstein et al., 1955; Bernstein and Ogata, 1966
Sunflowers	*Helianthus annuus* L.	4.8	2.7	5.0	MT	Cheng, 1983; Francois, 1996
Sugarbeets	*Beta vulgaris* L.	7.0	3.9	5.9	T	Bower et al., 1954
Wheat	*Triticum aestivum* L.	6.0	3.3	7.1	MT	Asana and Kale, 1965; Ayers et al., 1952; Hayward and Uhvits, 1944
Wheat, Durum	*T. turgidum* L. var. durum Desf.	5.9	3.3	3.8	T	Francois et al., 1986

† $EC_{1:1}$ is estimated based on a medium soil texture from Smith and Doran (1996)
‡ S = Sensitive, MS = Moderately Sensitive, MT = Moderately Tolerant, T = Tolerant (Maas and Hoffman, 1977)

THE INFLUENCE OF SOIL SALINITY GRADIENTS ON SOYBEAN [*GLYCINE MAX* (L.) MERR.] AND CORN (*ZEA MAYS* L.) GROWTH

Abstract

Salinization of agricultural lands is a worldwide resource concern greatly impacting agriculture production. In North Dakota, salinization has resulted from (1) a 25 year wet-cycle where naturally occurring salts from deep in the soil profile have been and continue to be transported to the surface and across the landscape and (2) reduced season-long water use as a result of crop selection. As a result, various levels of yield reduction are occurring, especially for salt-sensitive crops such as soybean [*Glycine max* (L.) Merr] and corn (*Zea Mays* L.). In this greenhouse study, the effects of salinity ($EC_{1:1}$ ranging from 0.40 (±0.06) to 4.2 (±0.45) dS m^{-1}) on above- and below-surface soybean parameters were evaluated. Corn productivity was evaluated on above- and below-surface measurements, across a salinity gradient ranging from $EC_{1:1}$ of 0.8 (±0.07) to 5.3 (±0.29) dS m^{-1}. The objective of this research was to determine soybean and corn response to salinity using methodology designed to resemble regional salinization, i.e. soluble salts accumulating in the root zone via capillary rise. From the control to the highest salinity; level leaf area (cm^2), plant mass (g), and plant height (cm) decreased by 66, 59, and 47%, respectively. Soybean root length (cm) and mass (g) were reduced by 47 and 29%, respectively; soil nitrate levels were also influenced significantly by increasing salinity. For corn above-ground parameters, crop mass, and height decreased from the control to highest $EC_{1:1}$ treatment by 42 and 26%, respectively, below-surface parameters, root length and mass also decreased across treatment levels, 44 and 37%, respectively. Findings from this study define the reduction in potential soybean and corn productivity for saline soils in the Northern Great Plains.

Statement of Objectives

The objective of this research was to define the relationship between soil salinity and the growth of salt sensitive crops, soybean and corn, for SO_4^- based soluble salts in northern Great Plains soils. For soybean, we hypothesized that (1) above-surface (leaf area, plant height and mass) and below-surface properties (root length and mass, nodulation) would decrease as soluble salt levels increase and (2) soil N pools, including ammonium (NH_4-N) and nitrate (NO_3-N), will increase with increasing salinity. For corn, we hypothesized that (3) above-surface (plant height and mass) and below-surface properties (root length and mass) would decrease as soluble salt levels increase.

Materials and Methods

Treatment Development

A greenhouse experiment was conducted in 2014 at the Agricultural Experiment Station Greenhouse Facility at North Dakota State University. The base material used for the experiment was a field-sourced loamy sand textured Glyndon (Coarse-silty, mixed, superactive, frigid Aeric Calciaquoll) soil collected near Hunter, North Dakota. The soil was air-dried, homogenized and sieved to 2 mm. The soil base was mixed in a 50:50 ratio with 2040 grade silica sand (TCC Materials, Mendota Heights, MN). A composite sample of this mixture was analyzed for soil physical and chemical properties at the North Dakota State University Soil Testing Laboratory (Table 3).

Table 3. Measured soil properties for a composite sample of Glyndon soil (Coarse-silty, mixed, superactive, frigid Aeric Calciaquolls) collected near Hunter, North Dakota mixed at a 50:50 ratio with silica quartz sand for greenhouse potting media.

Parameter	Analysis Result
pH	8.42
Electrical Conductivity	0.25 dS m^{-1}
Nitrate-N	51.2 mg kg^{-1}
Available P	30.9 mg kg^{-1}
Available K	118 mg kg^{-1}
Available Fe	1.80 mg kg^{-1}
Available Cu	0.57 mg kg^{-1}
Available Zn	1.34 mg kg^{-1}
Available Mn	4.10 mg kg^{-1}

The Glyndon soil (0.5 kg and 1 kg, for soybean and corn, respectively) and the silica sand (0.5 kg and 1 kg, for soybean and corn, respectively) were weighed separately. Nutrient solution was added directly to the silica sand for ease of mixing nutrients evenly. For soybean, P was added as K_2HPO_4 at a rate of 140.5 mg per pot. Micronutrient basal was added at a rate of 1 mg Fe, and 2.5 mg of each Zn, Mn and Cu; this was achieved with the additions of 11 mg $ZnSO_4 \bullet 7 H_2O$, 5.8 mg $MnSO_4 \bullet H_2O$, 6.3 mg $CuSO_4$. To reduce Fe chlorosis effects on soybean, FeEDDHA, 6% Fe (Soygreen granular, Sant Vicenc dels Horts Barcelona) was also applied at a rate of 0.3 mg per pot. Nutrient solution for corn consisting of 0.44 g of urea ($CO_2 (NH_4)_2$) and 0.44 g of KCl was added to the sand and mixed thoroughly.

After nutrients were applied to the sand, the soil was added to the sand and mixed thoroughly. Soil-sand mixes were split to allow for salt addition to half and reserve the other half. To ensure seed germination, one-half (0.5 kg and 1 kg for soybean and corn, respectively) of the soil-sand mixture was reserved. Soluble salts were added to one-half of the material. Treatments were established using soluble salts (Na_2SO_4 and $MgSO_4 \bullet 7H_2O$), added at four levels

0.0, 2.5, 5.8, and 8.8 g total salts, to artificially achieve a salinity gradient ranging from 0.40 to 4.2 dS m^{-1} (Table 4) for soybean and 0.75 to 5.3 dS m^{-1} (Table 5) for corn.

Table 4. Salinity levels achieved during soybean experiment showing the amount of soluble salts added per kg of potting media for an experiment conducted at the Agricultural Experiment Station Greenhouse Facility on the North Dakota State University main campus.

Target $EC_{1:1}$	Mass of salts added per kg of potting media		EC achieved from $EC_{1:1}$ extracts
	Na_2SO_4	$MgSO_4 \bullet 7H_2O$	
dS m^{-1}	------------g-------------		dS m^{-1}
0.3	0.0	0.0	0.40 (±0.1) †
2.0	1.3	1.2	1.7 (±0.3)
3.0	3.1	2.7	3.0 (±0.4)
4.0	4.7	4.1	4.2 (±0.5)

† Numbers within parentheses are standard deviation

Table 5. Salinity levels achieved during corn experiment showing the amount of soluble salt added per 2 kg container. The level achieved is represented by the Mean (±SD) for an experiment conducted at the Agricultural Experiment Station Greenhouse Facility on the North Dakota State University main campus.

Target $EC_{1:1}$	Mass of salts added per 2 kg of potting media		EC achieved from $EC_{1:1}$ extracts
	Na_2SO_4	$MgSO_4 \bullet 7H_2O$	
dS m^{-1}	------------g-------------		dS m^{-1}
0.3	0.0	0.0	0.75 (±0.1) †
2.0	2.7	2.3	2.50 (±0.4)
4.0	6.2	5.3	3.91 (±0.3)
5.0	9.4	8.1	5.34 (±0.3)

† Numbers within parentheses are standard deviation

The soil-sand mix treated with salt was then placed in a closed-bottom pot and topped with the reserved material, which contained no soluble salts (Figure 1). The same amount of salt was added for both soybean and corn, the difference in salinity level achieved for the two crops can be attributed to the different nutrients required for the respective crop, as nutrients are salts.

A control group was established, which contained no added soluble salts. Three seeds, for corn and soybean experiments, separately, were added and later thinned to one plant after emergence.

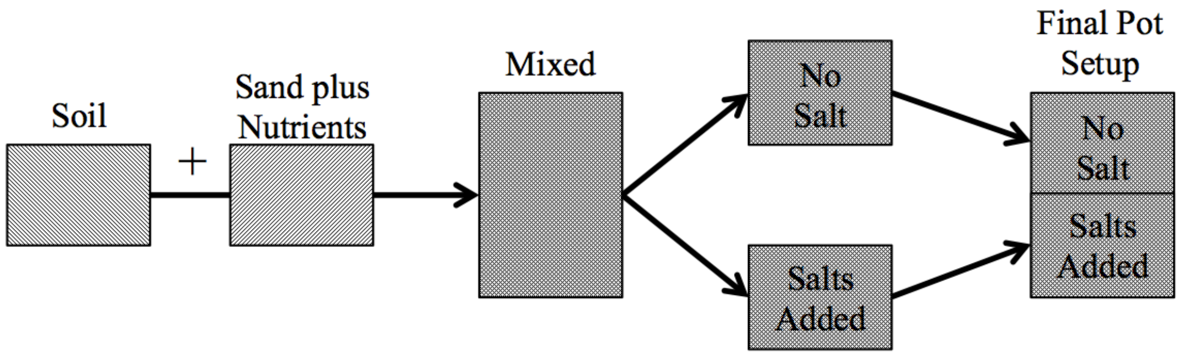

Figure 1. Methodology for pot setup for corn and soybean response to salinity experiment conducted at the Agricultural Experiment Station Greenhouse Facility on the North Dakota State University main campus.

Water (125 mL) was added to each pot to achieve 12% gravimetric water content for a loamy sand texture. Initial weights were recorded for gravimetric watering. Containers were watered to original gravimetric water weight every three days, initially. As plant water requirements increased, especially with corn, plants were watered daily. Containers were randomized on the benchtop in the greenhouse; replicates were rotated after watering. Growing conditions were 16 hr of light at 18.3-21.1°C and dark at 15.5-18.3°C for 8 hr. The experiment was completed in two blocks (n = 80 for each block), for both soybean and corn, with a total of 160 containers for each crop (n = 40 for each of the four targeted salinity levels).

Soil and Plant Analysis for Soybean

For the experiment, measurements [above- (leaf area, total mass, and height) and below-surface (root length and mass, NH_4-N, NO_3-N, and nodulation)] were completed at growth stages V1 and V2, i.e. 25 and 31 days of growth, respectively (n = 80 pots per time). Results from growth stages, V1 and V2, and experiment replications were analyzed together. Replication and growth stage were used as blocking variables to account for variation.

Leaf area was determined from the length and width of the central trifoliate leaf and height was measured from the soil surface to the tip of the central trifoliate leaf. For plant sampling, soybeans were cut at the stem within 1 cm of the soil surface, dried at 55°C, weighed, and ground (<0.1 mm). Volumetric root samples were collected using a 39 cm^3 core centered on the stem and pressed to the bottom of the pot. Root samples were air-dried, washed over a 2 mm sieve, and analyzed for length using a scanner and WinRHIZO software program (Regent Instruments, Quebec, QC). After scanning, roots were dried at 55°C, for 48 hours, weighed, and ground to pass through a 0.1 mm sieve.

Remaining soil from the container was air-dried, ground with a mortar and pestle to pass through a 2-mm sieve, air-dried and analyzed for electrical conductivity ($EC_{1:1}$) using a 1:1 soil to water ratio based on Rhoades (1996) using a conductivity meter (Model number VSTAR00, Versa Star Benchtop Meter, Swedesboro, NJ). Given the artificial mixture of sand and soil as a growth medium, use of the saturated pasted method (EC_e based on Rhoades (1996)) for determining soluble-salt levels produced highly variable results, as compared to $EC_{1:1}$, therefore the $EC_{1:1}$ method was used in this study. Soils were also analyzed for NO_3-N (Vendrell and Zupancic, 1990) and NH_4-N using flow injection (AutoAnalyzer 3 Seal AA3 Segmented Flow, Wet Chemistry Analyzer, Mequon, Wisconsin) (Markus et al. 1985). Plant, root and soil samples were analyzed for total N using high-temperature combustion (Elementar CN Analyzer, Hannau, Germany).

Soil and Plant Analysis for Corn

For the Corn experiment, measurements [above- (height and mass) and below-surface (root length and mass)] were completed at growth stages V6 and V8, 23 and 54 days of growth, respectively (n= 80 pots for each sampling period). Results from both stages and experiment replications were combined and analyzed; replication and growth stage were used as blocking variables to account for any variation.

Height measurements were taken from the surface of the growing medium to the top of the longest corn leaf. At both stages, corn plants were cut at the stem within 1 cm of the soil surface, dried at 55°C, weighed and ground (<0.1 mm). Volumetric root samples were also collected using a 5-cm diameter core, which was centered on the remaining stem fraction, removing a volume of 64 cm^3 of soil and roots. Samples were air-dried and roots were washed using a 2-mm sieve. Roots were analyzed using a scanner and WinRHIZO software program (Regent Instruments, Quebec, QC). Scanned roots were dried at 55°C and weighed.

Remaining soil from the container was air-dried and ground with a mortar and pestle to pass through a 2-mm sieve, and analyzed for $EC_{1:1}$ based on Rhoades (1996) using 10 g of soil: 10 mL deionized water mixture and measured with a conductivity meter (Versa Star Benchtop Meter, Swedesboro, NJ).

Statistical Analysis

Beyond plant response, the experiment also measured the effect of salinity on the reproduction of the two-spotted spider mite (*Tetranychus urticae* Koch) on both soybean and corn. Infestation was contained and isolated to a single leaf for a period of three days; due to the short span and isolation of two-spotted spider mite infestation we did not expect interference with overall plant response to salinity (J. Eichele, unpublished data, 2014).

Infestation and stage of growth were determined to have no significant influence on plant parameters, statistically. Regression analyses were used to determine the response of dependent soybean variables (leaf area, biomass, root length and mass and N concentrations) and corn variables (plant height, biomass, root length, and root mass) to the independent salinity levels ($EC_{1:1}$ dS m^{-1}) using JMP 11.1.1 (SAS Institute Inc., Cary, NC), SigmaPlot (Ver. 12, Systat Software Inc., San Jose, CA) was used for graph development. Separate regression equations were developed for each of the mentioned dependent variables for each crop. Significance was determined based on an alpha level of 0.05. The fit model in JMP 11.1.1 (SAS Institute Inc., Cary, NC) was used to account for any variation based on (1) the infestation of the two-spotted spider mite and (2) stage of plant from the two temporal blocks for both soybean and corn.

Results and Discussion

Soybean Response to Salinity

Leaf area of soybean decreased by 66% ($p < 0.0001$) as salinity treatment levels increased from $EC_{1:1}$ of 0.40 to 4.2 dS m^{-1} (Figure 2a). Leaf area was affected similarly in another study using NaCl salts at higher concentrations. Abel and MacKenzie (1964) noted a 90% decrease in leaf area at an estimated $EC_{1:1}$ of 7.7 dS m^{-1}, as compared to control (Table 1). Necrosis was severe on soybean leaves, at an $EC_{1:1}$ greater that 5.3 dS m^{-1}, in this 2013 experiment no leaf necrosis was noted. Leaves are the site of photosynthesis, thus, if leaf area is decreased so would the photosynthetic rate. Li et al. (2006) found that photosynthesis was adversely affected and transpiration rate declined for four soybean varieties as salinity increased from an estimated $EC_{1:1}$ of to 0.6 to 2.2 dS m^{-1}.

Soybean biomass decreased by 59% ($p < 0.0001$) as salinity treatment levels increased from $EC_{1:1}$ of 0.40 to 4.2 dS m^{-1} (Figure 2b). Decrease in biomass was also noted in other studies; however, methods using Cl-based salts showed lower declines in biomass at similar salinity levels (Table 1). Shalhevet et al. (1995) and van Hoorn et al. (2001) found a 21 and 39% decline in biomass for soybean, as compared to the 59% decline noted in this thesis study. Both studies used a natural soil and irrigated NaCl and $CaCl_2$ salt treatments, but differ in duration of the experiment. Shalhevet et al. (1995) conducted their experiment for two weeks, while van Hoorn et al. (2001) conducted the experiment to full soybean maturity. Therefore, the variation of soybean response across studies could be attributed to the salt type and salinization method.

Soybean variety many also influence biomass response to salinity. A previous study contrasted notable differences in salt tolerance among soybean varieties (Abel and MacKenzie, 1964). Other studies noted varietal differences for other legume crops, including pea. Shahid et al. (2012) found differences in salt tolerance among pea varieties. Tolerance ranged from low to high among 30 pea varieties exposed to a maximum estimated $EC_{1:1}$ of 5.6 dS m^{-1}, where decline in biomass ranged from 10 to 45% among varieties. Varietal differences in the salt tolerance of soybean and other legumes could lead to improved breeding lines for soybean salt tolerance.

Beyond reduction in above-ground biomass and leaf development, Ghassemi-Golezani and Taifeh-Noori (2011) found that the chlorophyll content index of soybean leaves was decreased with increasing salinity. Which reduces leaf functionality and exacerbates the photosynthesis-limiting factor of reduced leaf-surface area. Furthermore, Cl based salts are known to reduce stomata whereas SO_4 salts increase stomata numbers (Strogonov, 1962). This may also interfere with photosynthesis, plant water management, and overall plant productivity in saline environments. Ultimately, above-surface soybean parameters decline with increasing salinity, the rate of that decline is dependent on experimental methods.

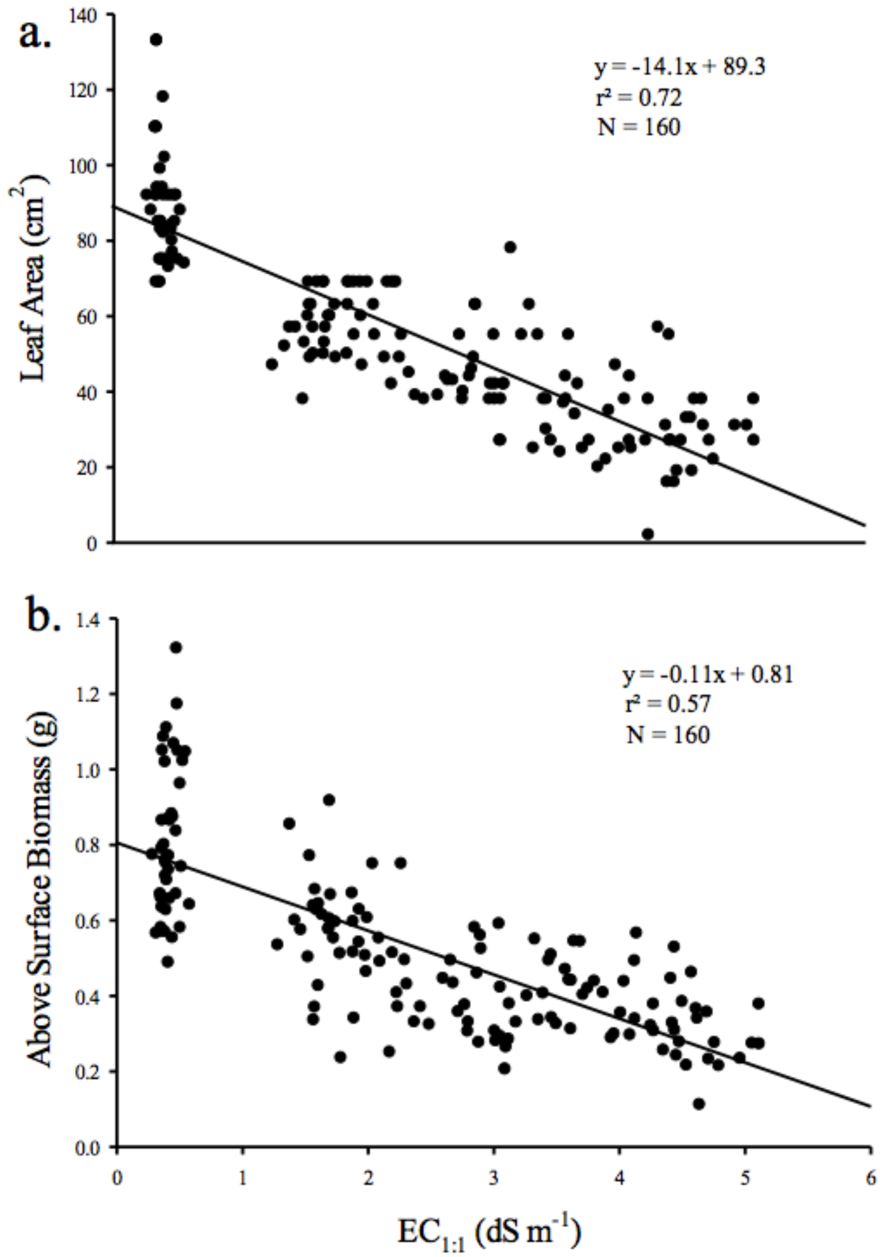

Figure 2. Regression line for a) soybean leaf area and b) soybean above-surface biomass against $EC_{1:1}$ (dS m^{-1}) for an experiment conducted at the Agricultural Experiment Station Greenhouse Facility on the North Dakota State University main campus

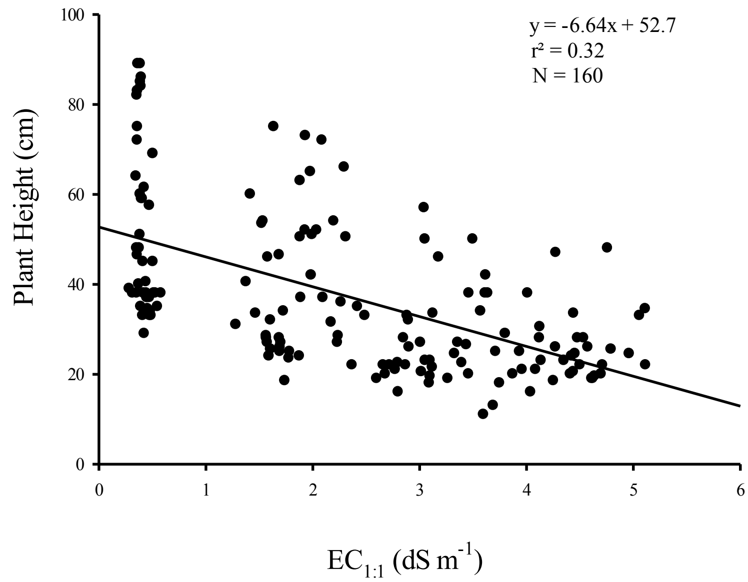

Figure 3. Regression line for soybean plant height in response to increasing $EC_{1:1}$ (dS m^{-1}) for an experiment conducted at the Agricultural Experiment Station Greenhouse Facility on the North Dakota State University main campus

Plant height (cm) also decreased significantly ($p < 0.0001$), by 47% as $EC_{1:1}$ salinity treatment levels increased from 0.40 to 4.2 dS m^{-1} (Figure 3). Shalhevet et al. (1995) noted a 29% difference in plant height over a period of two weeks, while Bustingorri and Lavado (2011) noted a 75% decline in plant height at early reproductive stage, R3. The 47% decline found in this thesis was between the other findings. Similarly, the 49% decline in height of the 'Manokin' soybean variety was determined by Wang and Shannon (1999) in a field irrigation study using NaCl and $CaCl_2$ salts. Clearly, there is a range in reported decline in plant height, which again, may be due to different methods and varieties used.

Yield data was not collected in this experiment; however, many studies have reported soybean yield response to salinity (Abel and MacKenzie, 1964; Hesterman, 1987; Maas, 1993; van Hoorn et al., 2001). For example Abel and MacKenzie (1964) examined salt tolerance during germination and production, finding that both were reduced as salinity increased. At the highest treatment level of 7.7 dS m^{-1}, $EC_{1:1}$, yield was negligible for four of the six varieties examined (Abel and MacKenzie, 1964). The threshold salinity tolerance level for soybean $EC_{1:1}$ has been estimated at 2.8 dS m^{-1}; beyond the threshold level relative yield is expected to decline at a 20% slope (Bernstein et al., 1955; Abel and MacKenzie, 1964; Bernstein and Ogata, 1966; Maas, 1993). Thus, there is a direct economic yield loss at levels of salinity greater than 2.8 dS m^{-1} ($EC_{1:1}$). The $EC_{1:1}$ is an estimate based on Smith and Doran (1996) where an EC_e equal to 5.0 dS m^{-1} would be an estimated $EC_{1:1}$ equal a 2.8 to dS m^{-1}, for a coarse to loamy sand.

Root length and mass decreased with increasing salinity (Figure 4a,b). Root length (cm) decreased by 47% from 0.40 to 4.2 dS m^{-1} and root mass (g) decreased by 29% across the increasing salinity gradient. The relationship between increased salinity and decreased root development and nodulation in soybean was also found in a number of other studies (Bernstein and Ogata, 1966; Singleton and Bohlool, 1984; Tu, 1981; Shalhevet et al., 1995; Wang and Shannon, 1999; Essa, 2002; Bustingorri and Lavado, 2011). Similar to previous greenhouse studies, root length declined by 31% in a field study by Wang and Shannon (1999) at an estimated $EC_{1:1}$ of 4.8 (Table 1). Osmotic and ionic mechanisms interfere with root development of salt sensitive crops (Bernstein, 1975), like soybean. When root length and mass significantly decrease on salt-stressed soybeans, water use is consequently decreased (Singleton and Bohlool, 1984).

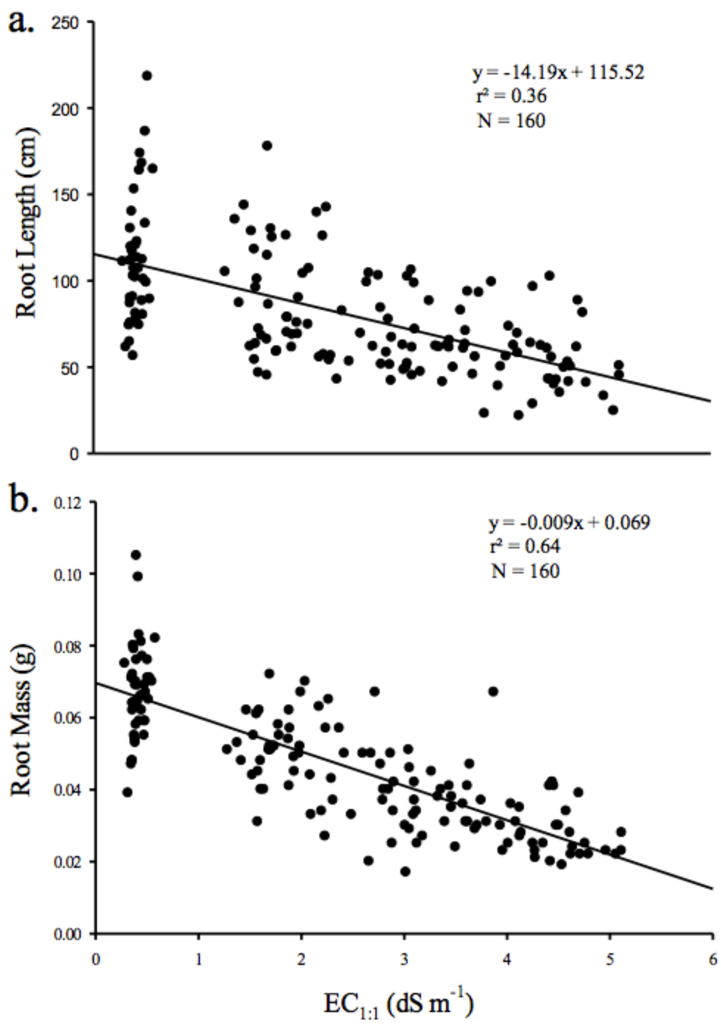

Figure 4. Regression line for a) soybean root length and b) soybean root mass against $EC_{1:1}$ (dS m^{-1}) for an experiment conducted at the Agricultural Experiment Station Greenhouse Facility on the North Dakota State University main campus

Reduction in nodulation has been correlated to root length and mass (Tu, 1981). This 2014 greenhouse thesis study, using sulfate based salts, found that nodulation decreased by 94% as salinity levels increased from 0.40 to 4.2 dS m^{-1} (Figure 5). Singleton and Bohlool (1984) used a split root experiment, half of the roots exposed to NaCl and half were not. The 1984 experiment found that NaCl adversely affects nodule initiation on roots exposed to salts; furthermore, timing of salinization impacted nodulation (Singleton and Bohlool, 1984).

Nodulation, in both number and mass, increased, as the time of salt treatment addition was delayed, i.e. salt applied at 0 hours contrasted to application at 96 hours (Singleton and Bohlool, 1984). Therefore, salts inhibit nodule formation, but after formation nodules can withstand low levels of salinity. Tu (1981) confirmed that NaCl salts resulted in significant reduction in nodulation with a solution of 0.8% NaCl additions and nodulation failed at a 1.2% NaCl application rate.

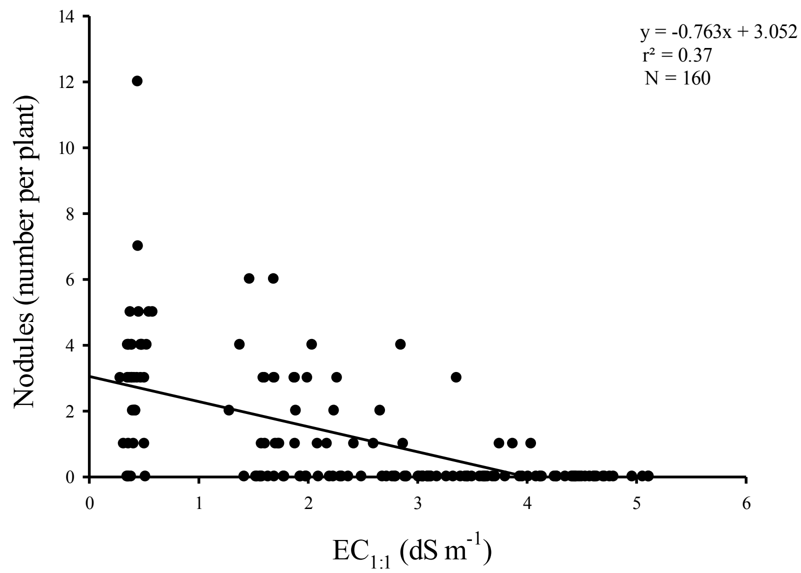

Figure 5. Regression line for soybean nodulation in response to increasing $EC_{1:1}$ (dS m^{-1}) for an experiment conducted at the Agricultural Experiment Station Greenhouse Facility on the North Dakota State University main campus

Soil NO_3-N values increased significantly ($p < 0.0001$) by 129% (8.33 to 19 mg kg^{-1}) as salinity increased from 0.40 to 4.2 dS m^{-1} (Figure 6a) Ammonium-N levels in the soil decreased, though not statistically significantly, by 27% as salinity levels increased from 0.40 to 4.2 dS m^{-1} (Figure 6b). Bernstein and Ogata (1966) and van Hoorn et al. (2001) showed similar results

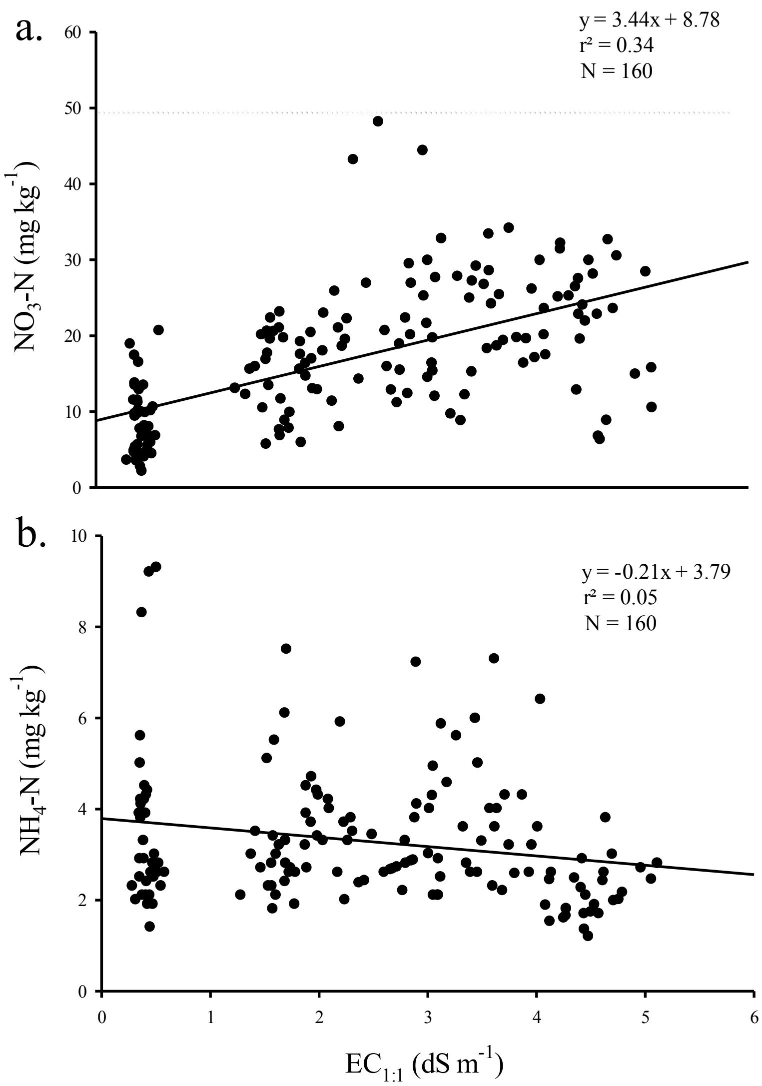

Figure 6. a) Nitrate-N (NO_3-N) and b) Ammonium-N (NH_4-N) in response to increasing soluble salt levels $EC_{1:1}$ (dS m^{-1}) for an experiment conducted at the Agricultural Experiment Station Greenhouse Facility on the North Dakota State University main campus

under different methodologies. Bernstein and Ogata (1966) measured NO_3-N and NH_4-N in soybean biomass and noted a significant increase in NH_4-N, and a numerical decrease in NO_3-N as salt levels increased to 5.4 atm. of NaCl. In this 2014 experiment soil NO_3-N increased and NH_4-N decreased significantly. Thus, the increase in NO_3-N and decrease in NH_4-N in the soil are reversed in the soybean dry material by Bernstein and Ogata (1966). Although NH_4-N increases in salt stressed plant material, NO_3-N accumulates in the soil. The increased NH_4-N accumulation in plant tissue found by Bernstein and Ogata (1966) may be in part due to the overall decrease in plant material and, thus, increased concentration of N. Studies focusing on other crops have found that excess nutrients will not compensate for salt stress (Bernstein, 1975). Total soil N pools (soil, root or plant tissue) were analyzed in this greenhouse study, no significant ($p < 0.0001$) differences were observed for any of the parameters. Van Hoorn et al. (2001) also found no relationship in N concentration and soil salinity, in shoots and pods of soybean and chickpea (*Cicer arietinum*), however, lentils (*Lens culinaris*) showed a decrease in plant material N concentrations as salinity increased. The 2001 study's treatment levels ranged from 0.5 to 3.9 dS m^{-1} ($EC_{1:1}$) using NaCl and $CaCl_2$ salts.

Nutrient cycling is important in the context of agricultural systems and soil salinity. In saline soils, crop yield will not be increased with the addition of nutrients including N (Bernstein, 1975); thus, soluble salts are the limiting factor for crop production on salt-affected soils. Sulfate salts impacted N uptake by soybean in this 2014 study. A previous study using Cl salts, with excess N applications, found that plant uptake of N decreased with increasing salinity (van Hoorn et al., 2001), resulting in N accumulation in the soil. Thus, moderately saline soils can accumulate inorganic N when continuously cropped with salt sensitive crops, i.e. soybeans and corn.

Combining the analyzed parameters of above- and below-surface plant growth and N cycling, the productivity of a salt sensitive crop, i.e. soybean, is limited under a gradient of increasing salinity levels. Water uptake by plants is reduced by the increased osmotic potential of the soil water containing soluble salts; at high concentrations most crops will exhibit drought-like symptoms (Munns and Tester, 2008). Over time crops can develop ion specific toxicity (Munns, 2002). Chloride, for example, can accumulate in the stems and leaves of soybean, disrupting cellular function (Abel and MacKenzie, 1964). Tolerant plant species, such as barley (*Hordeum vulgare* L.), are able to make osmotic adjustments with time to compensate for higher salinity levels; soybean has not been shown to make this adjustment as efficiently (Katerji et al., 2003). Although there is much research on soybean response to salinity, results are varied based primarily on method and potentially soybean variety; therefore, interpretation of results must consider the method used.

Corn Response to Salinity

Above-surface parameters, biomass and height, both decreased significantly ($p < 0.0001$), 42 and 26% as salinity treatment level increased from $EC_{1:1}$ 0.75 to 5.3 dS m^{-1}, respectively (Figure 7a,b). Studies using Cl based salts noted a range of decline in plant biomass as salinity increased, 14% (Shalhevet et al., 1995) and 22% (Bilgin et al., 2008) (Table 1). Hassan et al. (1970) experimented using a combination of SO_4 and Cl salts, noting a 98% decrease in biomass from the 0 dS m^{-1} to the highest salinity level of 8.7 dS m^{-1}, $EC_{1:1}$. Another study using irrigation salinization methods, found biomass yields were significantly decreased as $EC_{1:1}$ increased from 0.7 to 5.0 dS m^{-1} (Shani and Dudley, 2001). Plant height declined with increasing salinity by 36% (Shalhevet et al., 1995) and 8% (Bilgin et al., 2008) Shalhevet et al. (1995) noted higher declines in plant height (36%) compared to plant biomass (14%). Like Bilgin et al. (2008), this thesis study found lower declines in plant height compared to plant biomass. Shalhevet et al. (1995), Bilgin et al. (2008), and this thesis study analyzed the salinity response of the vegetative stage of corn. Compared to Shalhevet et al. (1995), Bilgin et al. (2008) and this thesis study focused on lower salinity levels compared to other studies. Corn height may be more affected at the higher salinity range compared to the lower end of the spectrum.

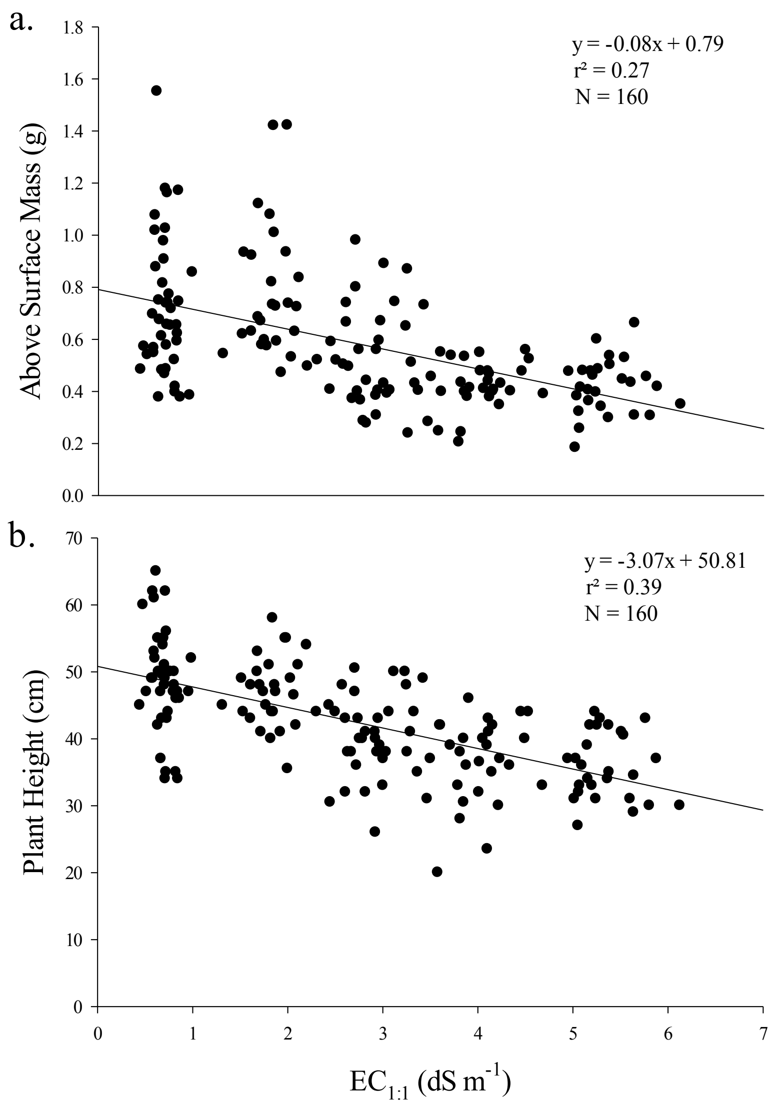

Figure 7. Regression line for a) corn above-surface mass and for b) plant height against $EC_{1:1}$ (dS m^{-1}) for an experiment conducted at the Agricultural Experiment Station Greenhouse Facility on the North Dakota State University main campus

Fewer studies focus on the below-surface plant parameters; typically the focus is placed on crop yield. This 2014 thesis experiment showed root length and mass decreased significantly ($p < 0.0001$) by 44 and 37%, as salinity treatment level increased from 0.75 to 5.3 dS m^{-1}, respectively (Figure 8a,b).

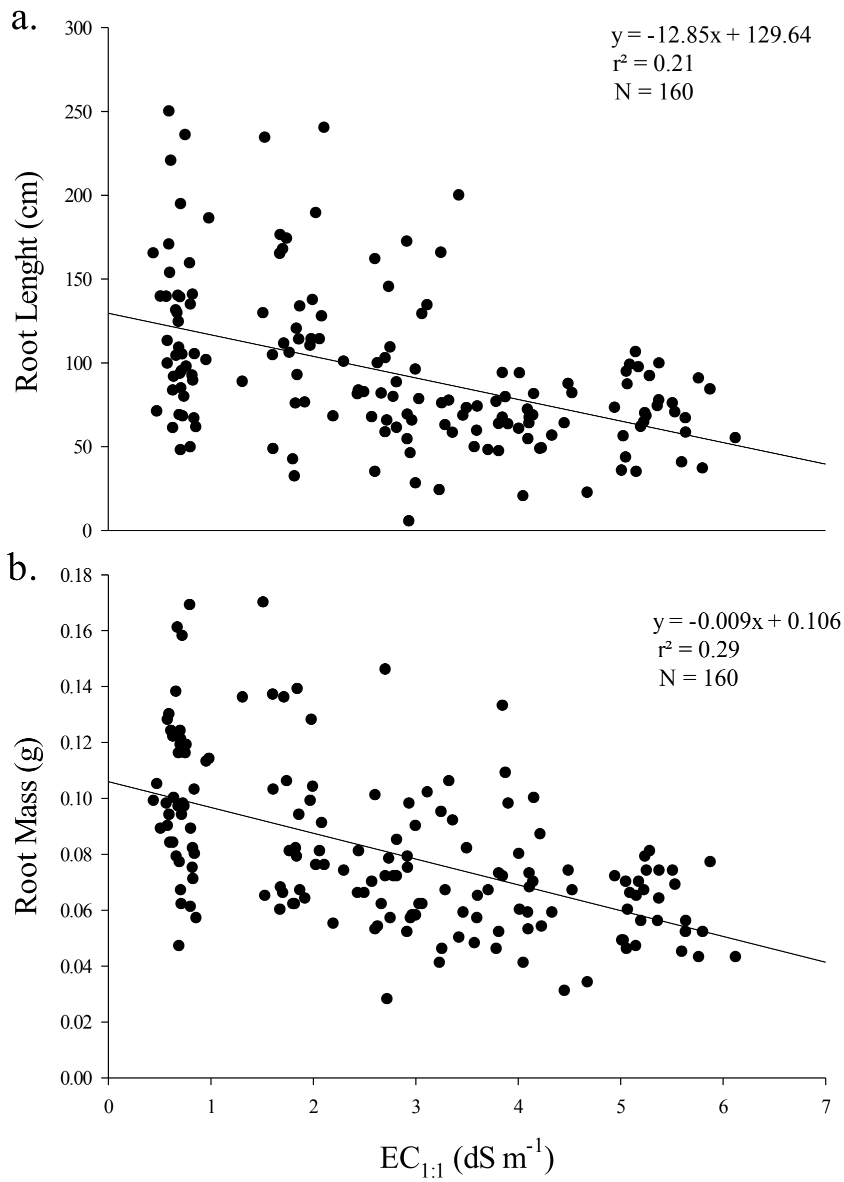

Figure 8. Regression line for below-surface parameters a) corn root length and for b) root mass $EC_{1:1}$ (dS m^{-1}) for an experiment conducted at the Agricultural Experiment Station Greenhouse Facility on the North Dakota State University main campus

A previous study by Bilgin et al. (2008), on corn response to salinity, found that below-surface parameters were less sensitive than shoot growth to salinity when using an NaCl irrigation solution increasing from 0 to 1.5 dS m^{-1}, $EC_{1:1}$. This response was not observed in this thesis study, plant height and root length decreased by 26 and 44%, respectively, as salinity increased. The discontinuity may be due to the SO_4-based salts used in this study, in comparison to the Cl-based salts used by Bilgin et al. (2008).

In general, saline soils have a decreased osmotic potential (Bernstein, 1975), where water and nutrient uptake is limited, inducing a drought-like stress. Shani and Dudley (2001) compared drought and salt stress in corn, finding that additional water does not compensate for a salt stress, furthermore, the amount of water required to reach maximum yield is decreased by increasing salinity. Limited water uptake in a saline soil system also results in limited nutrient uptake as well (Stewart et al., 1977; Katerji et al., 2003). However, Bernstein (1975) concluded that increased nutrient levels, including N, will not increase plant-salt tolerance; especially true at high salinity levels. Therefore, salinity, not fertility or water, is the limiting factor for corn production.

The entire corn plant, above and below-surface, is stunted due to salinity-induced stress. Although yield was not analyzed in this thesis experiment, yield has been the focus for many other experiments. Katerji et al. (2004) and Kaddah and Ghowail (1963) reported yield declines of 21 and 48%, respectively (Table 1) as salt levels increase. Corn is rated as a moderately sensitive crop with a salinity tolerance threshold level equal to an $EC_{1:1}$ of 1.0 dS m^{-1} (Bernstein and Ayers, 1949; Kaddah and Ghowail, 1964; Maas and Hoffman, 1977). Beyond an $EC_{1:1}$ of 1.0 dS m^{-1} corn productivity will decline at a slope of 12%, and thus, corn is not well suited for saline soils. However, a salt tolerant corn variety was released in 1987, Arizona 8601 (Day, 1987). The variety was an Indian maize source from a Native American reservation in Arizona and was reported to produce an average 26% more grain yield than a modern commercial variety in irrigated field trails (Day, 1987). Although Arizona 8601 is not a variety commonly in production, the release highlights the potential for corn production on saline soils. Although other corn varieties have not been released as salt tolerant, there is a range of tolerance in current varieties. For example out of the 19 hybrids studied by Eker et al. (2006), ‘Maverik’ was the most salt tolerant in terms of germination, biomass, and root production; furthermore, ‘Maverik’ had the lowest accumulation of Na in the leaf tissue. The variation in variety tolerance found by Eker et al. (2006) was vast, a 71% decline in biomass was found for the most sensitive variety, compared to a 52% decline in biomass for the most tolerant variety tested. The shoot concentration of Na demonstrates the amount of Na taken up by the plant from the soil; thus, if shoot or leaf Na concentration is high the plant is inefficient at Na exclusion and will eventfully lead to toxicity (Bernstein, 1975). Eker et al. (2006) found a 4-fold increase in shoot Na concentration in the sensitive to the more tolerant varieties. Thus, corn tolerance to salinity is related to the exclusion capacity of Na.

The decline in above- and below-surface parameters (plant height, plant mass, root length, and root mass) demonstrates that salinity using Na_2SO_4 and $MgSO_4 \cdot 7H_2O$ soluble salts negatively impacts all aspects of corn growth and development. There are distinct differences in the physiological responses of plants to SO_4 and Cl salts (Maas and Nieman, 1978); therefore, salinity tolerance levels for corn determined using chloride salts should be interpreted with that caveat. This 2014 greenhouse thesis experiment on corn response to regionally specific salt types and salinization defines that difference. Therefore, corn productivity, including above and below-surface variables, will be reduced on saline soils ranging from an $EC_{1:1}$ of 0.75 to 5.3 dS m^{-1} using SO_4 based salts.

Conclusion

Planting soybean and corn on salt-affected soils resulted in decreased plant productivity and has potential implications for salinity management beyond yield. Further research focusing on large-scale field analysis on the effects of soil salinity on above- and below-surface parameters and N cycling in the northern Great Plains region is an essential step to understanding the response of soybean and corn to low levels of salinity. Greenhouse experimentation allowed controlled and concise monitoring. Field research, although less controlled, will help define how soil salinity influences soybean and corn productivity and overall soil functionality in the northern Great Plains.

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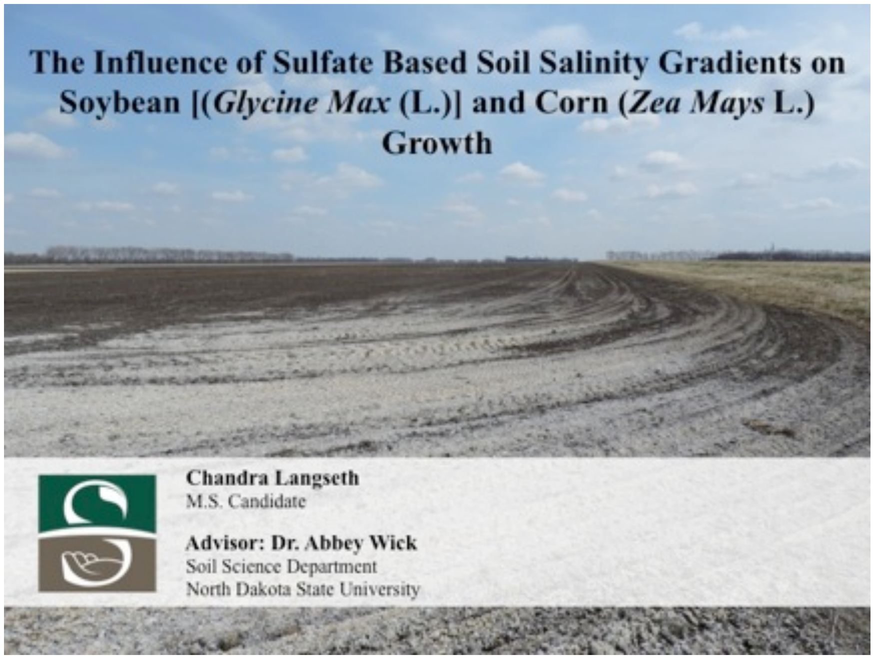
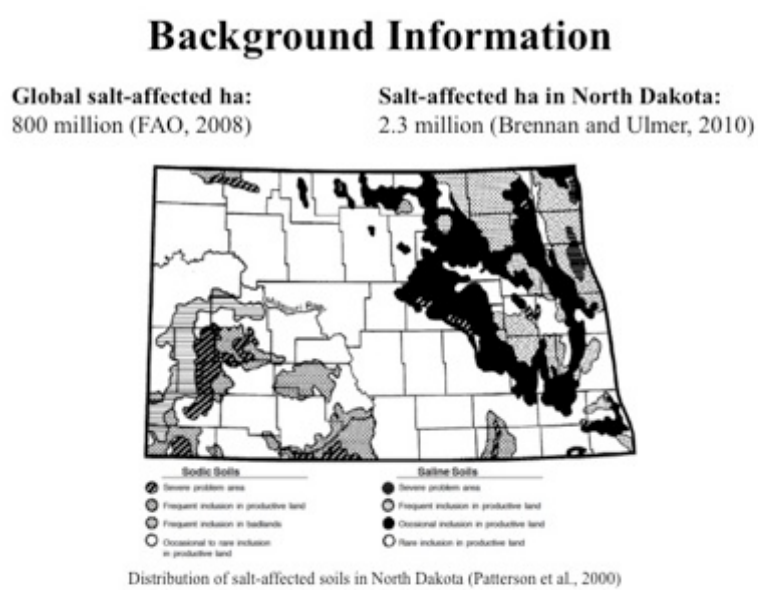
APPENDIX. SLIDE PRESENTATION





Background Information

Halophytes

- Greek root word *halo-* means "from salt"
- 2% of plants (Glenn and Brown, 1999)
- Complex salt tolerance mechanisms
  1. Ion exclusion
  2. Ion compartmentalization
  3. Ion excretion

Atriplex polycarpa, Saltbush

Glycophytes

- Greek root word *glycol-* meaning "sugar"
- Most crops
- Inefficient salt tolerance mechanisms
  1. Ion exclusion
  2. Ion compartmentalization

Glycine Max (L.) Merr, Soybean

Background Information

Salt Composition of ND Soils: $\mathbf{CaSO_4}$, $\mathbf{MgSO_4}$, $\mathbf{Na_2SO_4}$, and NaCl

Sulfate-Based Salts	Chloride-Based Salts
• Inhibits cell enlargement	• Enlargement of epidermal cells
• Decrease in succulence	• Decline in leaf surface
• Maintains cell division rates	• Limits cell division
• Increase in stomata	

(Maas and Nieman, 1978)

Background Information

Salinity tolerance for the major crops grown in North Dakota.

Crop	Scientific Name	Threshold $EC_{1:1}$†	Tolerance Rating‡	Reference
		dS m^{-1}		
Barley	*Hordeum vulgare* L.	4.5	**T**	Ayars et al., 1952; Hassan et al., 1970
Canola	*B. napus* L.	6.2	**T**	Francois, 1994
Corn	*Zea mays* L.	1.0	**MS**	Bernstein and Ayars, 1949; Kaddah and Ghowail, 1964
Dry Beans	-	0.6	**S**	Osawa, 1965
Flax	*Linum usitatissimum* L.	1.0	**MS**	Hayward and Spurr, 1944
Potatoes	*Solanum tuberosum* L.	0.8	**MS**	Bernstein et al., 1951
Soybean	*Glycine max* (L.) Merr.	2.8	**MT**	Abel and McKenzie, 1964; Bernstein et al., 1955; Bernstein and Ogata, 1966
Sunflowers	*Helianthus annuus* L.	2.7	**MT**	Cheng, 1983; Francois, 1996
Sugarbeets	*Beta vulgaris* L.	3.9	**T**	Bower et al., 1954
Wheat	*Triticum aestivum* L.	3.3	**MT**	Asana and Kale, 1965; Ayers et al., 1952; Hayward and Uhvits, 1944
Wheat, Durum	*T. turgidum* L. var. durum Desf.	3.3	**T**	Francois et al., 1986

† $EC_{1:1}$ is estimated based on a medium soil texture from Smith and Doran (1996)
‡ S = Sensitive, MS = Moderately Sensitive, MT = Moderately Tolerant, T = Tolerant (Maas and Hoffman, 1977)

Background Information

Compiled methodologies and results from previous soybean [*Glycine max* (L.) Merr.] and related crop salinity response greenhouse studies.

Crop	Salt Type	Method	$EC_{1:1}$ Range†	% Decline from low to high salinity treatment‡	Reference
			dS m^{-1}		
Soybean	NaCl	Greenhouse, irrigation	0.0 - 5.0	78% yield	Ghassemi-Golezani and Taifeh-Noori, 2011
Soybean	NaCl	Greenhouse	0.0 - 79.9 mM	- §	Singleton and Bohlool, 1984
Soybean	NaCl	Greenhouse	1.7 - 7.7	-	Abel and Mackenzie, 1964
Soybean	NaCl; $CaCl_2$	Greenhouse, irrigation	0.4 - 3.9	39% biomass; 46% yield	van Hoorn et al., 2001
Soybean	NaCl	Greenhouse, irrigation	0.0 - 1.6	-	Tu, 1981
Soybean	NaCl; $CaCl_2$	Greenhouse, irrigation	0.8 - 5.4	21% biomass; 29% plant height	Shalhevet et al., 1995
Soybean	NaCl	Greenhouse, irrigation	0.4 - 4.3	75% plant height	Bustingorri and Lavado, 2011
Soybean	NaCl	Greenhouse	0.0 - 5.4 atm	-	Bernstein and Ogata, 1966

† $EC_{1:1}$ is estimated based on a medium soil texture from Smith and Doran (1996)
‡ Estimated based on data reported in cited reference
§ Data not reported in reference

Background Information

Compiled methodologies and results from previous corn (*Zea Mays* L.) and related crop salinity response greenhouse studies.

Crop	Salt Type	Method	$EC_{1:1}$ Range†	% Decline from low to high salinity treatment‡	Reference
			$dS\ m^{-1}$		
Corn	NaCl	Greenhouse, irrigation	0.0 - 1.5	22% biomass 8% plant height	Bilgin et al., 2008
Corn	NaCl; $CaCl_2$	Greenhouse, irrigation	1.0 - 5.3	14% biomass 36% plant height	Shalhevet et al., 1995
Corn	Na_2SO_4; $MgSO_4$; $CaCl_2$	Greenhouse, irrigation	0.0 - 8.7	- §	Hassan et al., 1970
Corn	NaCl; $CaCl_2$	Field, irrigation	0.5 - 2.0	21% yield	Katerji et al., 2004
Corn	NaCl; $CaCl_2$	Field, irrigation	0.7 - 5.0	-	Shani and Dudley, 2001
Corn	NaCl; $CaCl_2$	Greenhouse, irrigation	0.2 - 5.0	-	Khalil et al., 1967
Corn	NaCl; $CaCl_2$	Field, irrigation	0.3 - 4.2	-	Shenker et al., 2003
Corn	NaCl; $CaCl_2$	Field, irrigation	0.0 - 7.9	48% yield	Kaddah and Ghowail, 1963

† $EC_{1:1}$ is estimated based on a medium soil texture from Smith and Doran (1996)
‡ Estimated based on data reported in cited reference
§ Data not reported in reference

Objectives

The objective of this research is to determine soybean and corn response to salinity using methodology designed to resemble regional salinization, i.e. sulfate based salts accumulating in the root zone via capillary rise.

Hypothesis

For **soybean**, I hypothesized that:

1. Above- and below- surface properties will *decrease* as soluble salt levels increase
2. Soil N pools, including ammonium (NH_4-N) and nitrate (NO_3-N), will *increase* with increasing salinity.

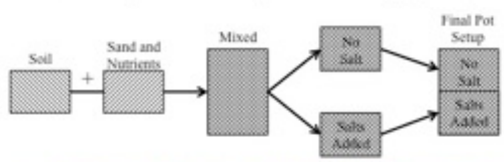
For **corn**, I hypothesize that:

1. Above- and below- surface properties will *decrease* as soluble salt levels increase

Experimental Setup

50%: Field-sourced Glyndon (Coarse-silty, mixed, superactive, frigid Aeric Calciaquoll)

50%: 2040 grade silica sand (TCC Materials, Mendota Heights, MN)



Salinity levels achieved during **soybean** experiment

Target $EC_{1:1}$	Mass of salts added per kg of potting media		EC achieved from $EC_{1:1}$
	Na_2SO_4	$MgSO_4 \cdot 7H_2O$	
dS m^{-1}	------------g-------------		dS m^{-1}
0.33	0.00	0.00	**0.40 (±0.1)** †
2.0	1.3	1.2	**1.7 (±0.3)**
3.0	3.1	2.7	**3.0 (±0.4)**
4.0	4.7	4.1	**4.2 (±0.5)**

† Numbers within parentheses are standard deviation

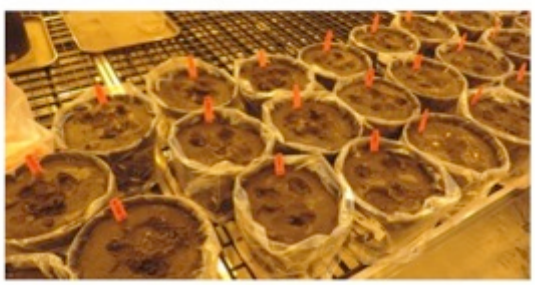
Salinity levels achieved during **corn** experiment

Target $EC_{1:1}$	Mass of salts added per 2 kg of potting media		EC achieved from $EC_{1:1}$
	Na_2SO_4	$MgSO_4 \cdot 7H_2O$	
dS m^{-1}	------------g-------------		dS m^{-1}
0.33	0.00	0.00	**0.75 (±0.1)** †
2.0	2.7	2.3	**2.50 (±0.4)**
4.0	6.2	5.3	**3.91 (±0.3)**
5.0	9.4	8.1	**5.34 (±0.3)**

† Numbers within parentheses are standard deviation

Experimental Setup

- Four seeds per pot, later thinned to one
- Gravimetrically watered to 12% gravimetric water content for a loamy sand



- Two temporal blocks were set up for each crop
- A total of 160 pots per crop
  - 40 pots per treatment

Plant Sampling

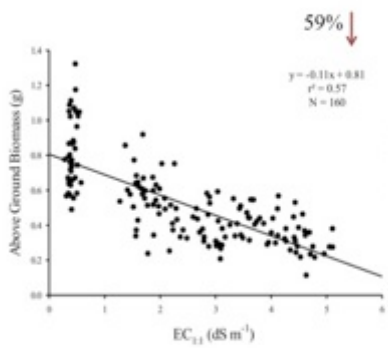
Corn	Soybean
V6 and V8	V1 and V2
Biomass cut at surface	Biomass cut at surface
64 cm^3 root core	39 cm^3 root core

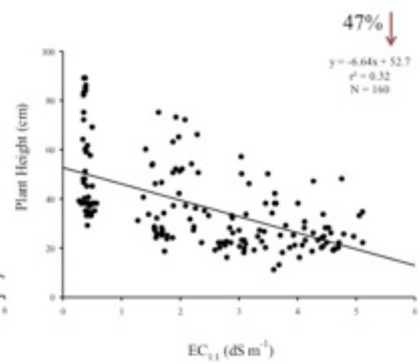
Soil Analysis

- Soil was air dried, ground with mortar and pestle
  - $EC_{1:1}$ method was used (Rhoades, 1996)

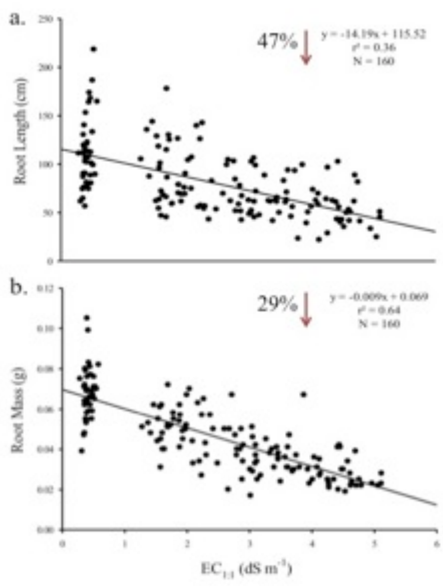
Study 1. Soybean Response

Study 1. Soybean Response





Study 1. Soybean Response



Root scan at 0.4 dS m^{-1}

Study 1. Soybean Response

Crop	Salt Type	Method	$EC_{1:1}$ Range†	% Decline from low to high salinity treatment‡	Reference
			$dS\ m^{-1}$		
Soybean	Na_2SO_4; $MgSO_4 \cdot 7H_2O$	Greenhouse, dryland	0.4 - 4.2	59% biomass 47% plant height	Langseth et al., 2014
Soybean	NaCl	Greenhouse, irrigation	0.0 - 5.0	78% yield	Ghassemi-Golezani and Taifeh-Noori, 2011
Soybean	NaCl; $CaCl_2$	Greenhouse, irrigation	0.4 - 3.9	39% biomass 46% yield	van Hoorn et al., 2001
Soybean	NaCl; $CaCl_2$	Greenhouse, irrigation	0.8 - 5.4	21% biomass 29% plant height	Shalhevet et al., 1995
Soybean	NaCl	Greenhouse, irrigation	0.4 - 4.3	75% plant height	Bustingorri and Lavado, 2011

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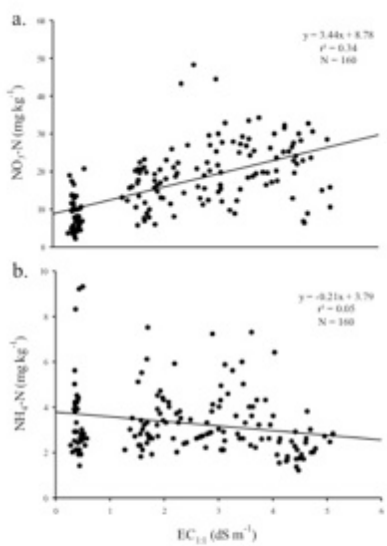

Study 2. Corn Response

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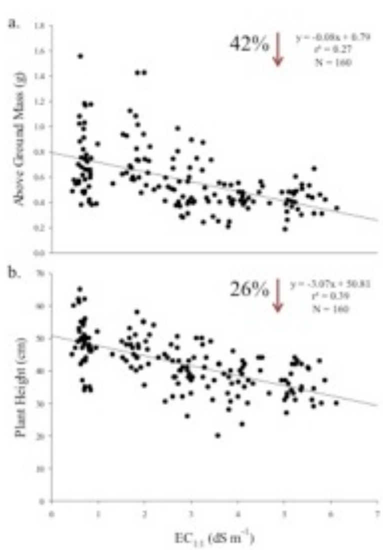
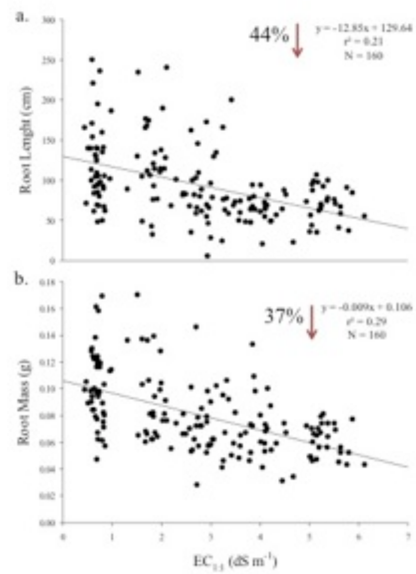


Study 2. Corn Response



Root scan at 0.8 dS m^{-1}

Study 2. Corn Response

Crop	Salt Type	Method	$EC_{1:1}$ Range†	% Decline from low to high salinity treatment‡	Reference
			dS m^{-1}		
Corn	Na_2SO_4; $MgSO_4 \cdot 7H_2O$	Greenhouse, dryland	0.8 - 5.3	42% biomass 26% plant height	Langseth et al., 2014
Corn	NaCl	Greenhouse, irrigation	0.0 - 1.5	22% biomass 8% plant height	Bilgin et al., 2008
Corn	NaCl; $CaCl_2$	Greenhouse, irrigation	1.0 - 5.3	14% biomass 36% plant height	Shalhevet et al., 1995
Corn	NaCl; $CaCl_2$	Field, irrigation	0.5 - 2.0	21% yield	Katerji et al., 2004
Corn	NaCl; $CaCl_2$	Field, irrigation	0.0 - 7.9	48% yield	Kaddah and Ghowail, 1963

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Corn	NaCl; $CaCl_2$	Field, irrigation	0.5 - 2.0	21% yield	Katerji et al., 2004
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Conclusion

Planting soybean and corn on salt affected soil results in:

- Decreased productivity, above- and below-surface
- Increase in soil nitrate

Greenhouse experimentation is the first step in determining soybean and corn response to regional salinity. Field research on crop response to salinity is the next step.

Questions?

Chandra Langseth, M.S. Candidate

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Committee Members:
Dr. Thomas DeSutter
Dr. Jason Harmon

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North Dakota Soybean Council
Our World Is Growing.

North Dakota Corn Council

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