

## ABSTRACT

BESELI, AMBER LYNN. Advancing Understanding of Climate Change: Undergraduate Education and Maize Drought Response. (Under the direction of Dr. Thomas R. Sinclair).

As the researcher sought to earn a PhD in Crop Science with a Minor in Agricultural Education, this PhD dissertation follows a unique approach in that two chapters are based on plant science research and one chapter is based on educational research. The common thread that ties these two types of research together is climate change. The plant-science research chapters (Chapters 2 and 3) focus on improved drought tolerance in maize. The educational research chapter (Chapter 4) focuses on how university professors teach undergraduates about climate change.

The research presented in Chapters 2 and 3 of this dissertation involved two lines of maize (*Zea mays* L.) that have been previously studied by Chimenti, Marcantonio, and Hall (2006) – a high osmotic adjustment line (HOA) and a low osmotic adjustment line (LOA). The studies in Chapter 2 of this dissertation examined these two lines of maize in further detail and aimed to find possible reasons for HOA lines' delayed wilting phenotype. In Chapter 2, neither the lack of differences in osmotic potential in the root tips, nor the lack of differences in rooting length, could explain observed differences in wilting between the two lines under drought. Because of that an alternative hypothesis related to difference in the water use by the two lines over time was explored in Chapter 3 of this dissertation.

In Chapter 3, it was discovered that the two lines of maize respond quite differently to vapor pressure deficit. Under low VPD conditions and in the early part of the dry-down cycle, the HOA Line had greater transpiration rates than the LOA Line. This observed difference in transpiration rate is consistent with the lower leaf OP for the HOA Line (results from Chapter 2) under low VPD conditions resulting in a greater hydrostatic potential gradient between the soil

and leaves in the HOA Line as compared to the LOA Line. The visual consequences of higher hydrostatic pressure in the HOA Line would be delayed wilting. The results of the experiments in Chapter 3 showed that the higher transpiration early in the drying cycle caused the water to be used more rapidly and the HOA Line reached the endpoint in the use of transpirable soil water earlier than LOA. As the findings of Chapters 2 and 3 show, these two lines of maize – the HOA Line and the LOA Line – do not appear to have outstanding drought tolerant mechanisms at this current point in time. However, these two lines have been useful in expanding our understanding of how osmotic potential impacts the rate of water uptake.

The research presented in Chapter 4 of this dissertation focused on how university professors teach undergraduates about climate change. The population of this descriptive quantitative research study was comprised of North Carolina State University faculty who have taught an undergraduate course that contained one of the following phrases in their course description: “climate change,” “global warming,” or “environmental impact.” Twenty-four professors completed a cross-sectional survey questionnaire that sought to understand how they teach about climate change. The specific research objectives of this chapter were to: (1) understand professors’ views on climate change, (2) explore which climate change-related content they teach in their courses, and (3) understand which teaching methods they use to teach undergraduates about climate change. Results from this study indicate that professors believe climate change is occurring, but they do not share a clear consensus of the cause of climate change. Local and state policy related to climate change is lacking from climate change curriculum. This study adds to the literature on climate change education in the university.

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Advancing Understanding of Climate Change: Undergraduate Education and Maize Drought  
Response

by  
Amber Lynn Beseli

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North Carolina State University  
in partial fulfillment of the  
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## **DEDICATION**

To my grandparents, Billy and Debbie Hoyle, better known as Meme and Pepaw. Thank you for all you do for our family and me.

## **BIOGRAPHY**

Amber Lynn (Willis) Beşeli was born and raised in Belwood, North Carolina to her parents Steve and Angie Weathers. Amber's passion for agricultural education formed while she was a student at Burns High School, under the direction of Mr. Luke Beam and Mr. Dennis Martin. After graduating from Burns in 2009, Amber moved to Raleigh, NC to attend NC State. In December 2012, Amber graduated Magna Cum Laude with her Bachelor of Science degree in Agricultural Education with a minor and concentration in Horticultural Science, under the advisement of Dr. Beth Wilson. In August 2014, Amber graduated with her Master of Science degree in Agricultural and Extension Education, under the direction of Dr. Wendy Warner.

In July 2013, Amber began working in Dr. Tom Sinclair's research group, assisting with drought tolerance experiments. After a growing interest and appreciation in this work, Amber began her Crop Science PhD program in August 2014, under the direction of Dr. Sinclair.

Throughout her time at NC State, Amber had several opportunities to study abroad and help lead study abroad programs. Through these experiences, Amber's love for international experiences, international agriculture education, and other cultures have continued to deepen. In 2015, Amber created a new Agricultural Institute course, Global Issues in Agriculture, which she has taught each spring semester. In May 2017, Amber started working in NC State CALS in International Programs as the Program Coordinator for CALS Global Academy. After graduating from her PhD program, Amber will continue to work for CALS International Programs.

While in graduate school, Amber met Aydin Beşeli of Istanbul, Turkey and they married in 2015. In her spare time, Amber loves traveling the world with her husband and spending time at Lake James. When not traveling, you can find Amber daydreaming of Prague and Guatemala.

## ACKNOWLEDGMENTS

As anyone who has completed a graduate program knows, there are many people behind the graduate student helping them every step along the way. I have many people to thank for helping me get to where I am today.

First of all I would like to thank my PhD committee for all of their time, help, and support.

Tom, thank you for giving me the opportunity to work in your lab and then for giving me the chance to start my own PhD program! You are an excellent mentor – always explaining things, always here to talk, and even open for science questions on your home phone! Thank you for all of your guidance – I would not have wished to be brainwashed by anyone else!

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## TABLE OF CONTENTS

<b>LIST OF TABLES</b> .....	xi
<b>LIST OF FIGURES</b> .....	xii
<b>CHAPTER 1: Introduction</b> .....	1
Climate Change: The Common Thread .....	1
Improved Drought Tolerance in Maize.....	3
Climate Change Education .....	8
Dissertation Overview .....	10
<b>CHAPTER 2: Comparing Rooting Lengths and Root Osmotic Potential Between Two Lines of Maize</b> .....	13
Introduction.....	13
Materials and Methods.....	15
Results.....	25
Discussion.....	27
<b>CHAPTER 3: Dynamics of Temporal Water Use in Two Lines of Maize</b> .....	40
Introduction.....	40
Materials and Methods.....	42
Results.....	50
Discussion.....	52
<b>CHAPTER 4: Understanding How University Professors Teach about Climate Change...</b>	62
Introduction.....	62
Methodology .....	67
Results.....	75
Discussion.....	96
<b>CHAPTER 5: Final Discussion, Recommendations, and Implications</b> .....	104
Introduction.....	104
Chapter 2 – Comparing Rooting Lengths and Root Osmotic Potential Between Two Lines of Maize .....	105
Chapter 3 – Dynamics of Temporal Water Use in Two Lines of Maize .....	106
Chapter 4 – How University Professors Teach About Climate Change.....	109
Final Thoughts .....	110
<b>REFERENCES</b> .....	112
Chapter 1 References .....	112
Chapter 2 References .....	117

Chapter 3 References .....	118
Chapter 4 References .....	119
Chapter 5 References .....	121
<b>APPENDICES</b> .....	122
Appendix A .....	123
Appendix B .....	124
Appendix C .....	125
Appendix D .....	126
Appendix E .....	127
Appendix F .....	128
Appendix G .....	144

## LIST OF TABLES

Table 2.1	Overview of each experiment with their environmental conditions and important dates.....	39
Table 4.1	Reliability Statistics for All Three Constructs .....	69
Table 4.2	Comparing Early and Late Respondents with an Independent Samples T-Test .....	72
Table 4.3	Descriptive Statistics for the Professors' Assumptions of What Their Students Know About Climate Change Prior to Their Class .....	77
Table 4.4	Descriptive Statistics for Impacts of Climate Change Content .....	80
Table 4.5	Descriptive Statistics for Greenhouse Effect Content and Policy Content .....	82
Table 4.6	Correlation matrix for Impacts of Climate Change Content .....	85
Table 4.7	Correlation matrix for Greenhouse Effect and Policy Content. ....	86
Table 4.8	Correlation matrix for Impacts of Climate Change Content and Assumptions about Student Knowledge Prior to the Professor's Course .....	88
Table 4.9	Correlation matrix for Greenhouse Effect and Policy with Assumptions about Student Knowledge Prior to the Professor's Course .....	89
Table 4.10	Correlation matrix for Percentage of Students per College and Percentage of Each Year of Undergraduate Studies with Assumptions about Student Knowledge Prior to the Professor's Course .....	90
Table 4.11	Descriptive Statistics for Teaching Methods Used to Teach Climate Change.....	92
Table 4.12	Correlation matrix for Teaching Methods.....	94
Table 4.13	Descriptive Statistics for Recourses Used to Help Professors Gather Information to Teach About Climate Change .....	95

## LIST OF FIGURES

Figure 1.1	Regional patterns of yield variation due to change in rooting .....	12
Figure 2.1	GC-LVPD: Leaf Osmotic Potential vs. NTR. ....	32
Figure 2.2	GC-LVPD: Nodal Root Tip Osmotic Potential vs. NTR.. ....	33
Figure 2.3	GC-MVPD: Comparing the Average Nodal Root Lengths of the HOA and LOA Lines for Each Harvest Day .....	34
Figure 2.4	GC-HVPD: Comparing the Average Nodal Root Lengths of the HOA and LOA Lines for Each Harvest... ..	35
Figure 2.5	GC-LVPD: Comparing the Average Nodal Root Lengths of the HOA and LOA Lines for Each Harvest... ..	36
Figure 2.6	FES: Comparing the Average Root Lengths of the HOA and LOA Lines for Each Harvest.....	37
Figure 2.7	FLS: Comparing the Average Root Lengths of the HOA and LOA Lines for Each Harvest.....	38
Figure 3.1	Transpiration rate under well-watered conditions of the LOA Line and the HOA Line exposed to increasing VPD levels under controlled conditions in a growth chamber .....	57
Figure 3.2	Exponential model was fit for normalized transpiration rate (NTR) versus fraction of transpirable soil water (FTSW) for the LOA Line and the HOA Line... ..	58
Figure 3.3	Exponential model was fit for normalized transpiration rate (NTR) versus fraction of transpirable soil water (FTSW) for the LOA Line and the HOA Line... ..	59
Figure 3.4	GC-LVPD: NTR vs. FTSW. ....	60
Figure 3.5	Difference in Average Water Use between the HOA Line and LOA Line.....	61
Figure 4.1	Proposed Theoretical Framework for Teachers' Attitudes toward (the Teaching) of Science .....	66

## **CHAPTER 1: Introduction**

### **Climate Change: The Common Thread**

“There’s one issue that will define the contours of this century more dramatically than any other, and that is the urgent threat of a changing climate.” These words of U.S. President Barack Obama are echoed by scientists, researchers, political leaders, and religious leaders across the globe. As climate change is considered one of the largest challenges facing our current and future generations, specifically related to food production (Battisti and Naylor, 2009), many people in science and policy are urgently working to adapt to climate change.

Effects of climate change include temperature rise, lengthening of growing seasons and frost-free seasons, changes in precipitation patterns, stronger hurricanes, sea-level rise, melting of sea ice in the Arctic, and more droughts and heat waves (U.S. Global Change Research Program, 2014). Each of these effects of climate change have an impact on agriculture, and as the climate changes more intensely, the effects, such as drought, will worsen.

Not only is agriculture impacted by climate change, but agriculture also has a large impact on climate change (Rosenzweig, 2011). Agricultural production is a major source of greenhouse gas emissions, such as carbon dioxide (CO<sub>2</sub>), methane (CH<sub>4</sub>), and nitrous oxide (N<sub>2</sub>O) (Cole et al., 1997; IPCC, 2001). Decay of and burning of plant litter and soil organic matter, land-use change, and farming operations release large amounts of carbon dioxide into the atmosphere (Janzen, 2004; Rosenzweig, 2011; Smith, Smith, and Conen, 2004). Flooded rice production, ruminant livestock digestion, and manure lagoons allow methane release (Mosier et al., 1998; Rosenzweig, 2011). Nitrous oxide is largely emitted through excess nitrogen fertilizer application (Rosenzweig, 2011; K. A. Smith et al., 2004).

In order to adapt to the changing climate, and in order to reduce our greenhouse emissions, the society must become more educated on climate change and climate science. As Smith et al. (2008) wrote, “The gap between technical potential and realized GHG mitigation occurs due to barriers to implementation, including climate and non-climate policy, and institutional, social, educational and economic constraints.” This PhD dissertation focuses on how to both adapt to climate change in crop production and how to educate the younger generation about climate change and climate science.

As the researcher sought to earn a PhD in Crop Science with a Minor in Agricultural Education, this PhD dissertation follows a unique approach in that two chapters are based on plant science research and one chapter is based on educational research. The common thread that ties these two types of research together is climate change. The plant science research chapters (Chapters 2 and 3) focus on improved drought tolerance in maize. Droughts increase and water patterns change with climate change, hence the increasing need for drought tolerant crops. The educational research chapter (Chapter 4) focuses on how university professors teach undergraduates about climate change. As climate change is one of the largest threats to our livelihoods, university students should be educated about this scientific topic.

### **Definition of Key Terms**

The term climate change used throughout this dissertation is based off the definition from the Intergovernmental Panel on Climate Change (IPCC). According to the IPCC (2007), climate change is

A change in the state of the climate that can be identified (e.g. using statistical tests) by changes in the mean and/or the variability of its properties, and that persists for an

extended period, typically decades or longer. It refers to any change in climate over time, whether due to natural variability or as a result of human activity.

There are many differing definitions of climate change but this one was most appropriate for this research because not just one possible cause was named; the IPCC definition mentioned both human influence and natural causes in their definition.

### **Improved Drought Tolerance in Maize**

One major challenge to increasing crop yields in many parts of the world is drought (Sinclair, 2017). A crop experiences drought when an imbalance occurs in their water supply needs, specifically when the rate of water supply to the leaves is insufficient to keep up with their transpiration rate. When the plant's need for water is not met, the plant's water content decreases (Blum, 2005). For a crop to be considered drought tolerant, the plant and its cells must maintain physiological activity, even when there are only low levels of water available (Sinclair and Weiss, 2010). A plant can achieve drought tolerance by two approaches: an ability to survive dehydration or mechanisms to avoid dehydration in the first place.

Approaches simply to enhance crop survival are likely not issues in many crop plants, especially in annual crop plants. This is because, if the crop plants do survive a period of severe drought, they will still probably yield very little, and therefore bring low profits to the farmer. That is, even if the crop plants survive the drought, they will still not have enough water needed to support high quality growth and high yields (Sinclair, 2000). Although, crop survival through drought is important in perennial plants, such as forages (Sinclair, 2000).

If a plant is capable of avoiding dehydration, then it is able to keep water in the plant or keep their cells hydrated during the period of soil water deficit. If plants avoid dehydration then

they are not stressed because they do not allow their tissues to become dry (Blum, 2005). For example, root extension of 5 mm per day can sustain the crop plants at the beginning stage of severe drought if the leaf area index and/or epidermal conductance are small by allowing the crop to continue daily water uptake (Sinclair, 2000). A corollary of sustained root extension is that the plants are likely to achieve deeper rooting. Many years ago, Cannon (1949) found that the plants showing the most drought tolerance had deeper root systems.

### **Osmotic Adjustment and Root Extension for Improved Drought Tolerance**

One important drought tolerance mechanism claimed for crops is osmotic adjustment (Blum, 1989; Blum, 2017; Hare et al., 1998; Moinuddin and Khanna-Chopra, 2004). Osmotic adjustment (OA) is a plant adaptive response that occurs at the cellular level (Blum, 2005) that is often assumed to be an important drought tolerant mechanism. Osmotic adjustment is when the osmotic potential decreases because solutes concentration in cells increase. During osmotic adjustment, plant cells can gather solutes and this has two consequences. One consequence of increased solute concentration is that the tissue water potential is likely to be decreased. Lowering of the water potential is crucial because plants can uptake water if the water potential is lower, i.e., more negative, than the soil water potential (Taiz, Zeiger, Møller, and Murphy, 2015). Osmotic adjustment in the roots in particular could improve root capacity for water uptake (Chimenti, Marcantonio, and Hall, 2006; Tangpremsri, Fukai, Fischer, and Henzell, 1991).

A second consequence of osmotic adjustment can be an infusion of water in the cells that result in a higher tissue turgor pressure. If turgor pressure is maintained or even increased in root tips, then continued root extension into drying soil seems possible (Sharp and Davies, 1989).

Osmotic adjustment has been reported in the tips of maize nodal and seminal roots (Sharp & Davies, 1979; Westgate & Boyer, 1985). Nodal root apices of maize have the ability to osmotically adjust and retain high turgor when there is low soil water potential (Sharp & Davies, 1979). In sorghum, Wright and Smith (1983) found that more water was extracted from deeper in the soil profile because likely due to osmotic adjustment.

Blum (2005) wrote, “At the end of the day the essence of the matter is that where deep soil moisture is available a long root to reach this moisture is simply as effective as a long rope in a deep well.” There are often useful amounts of water down in the subsoil under plants experiencing drought stress (Passioura, 1985). Passioura (2007) pointed out that in regard to adjusting plant phenotypic traits to better withstand drought-stress of cereal crops, the greatest potential may lay in increasing root penetration and depth in the soil. However, a crop plant with deeper rooting may exhaust the water stored deeply and this water is not readily recharged. Once plants deplete the soil water reserves, the crop would be worse off than before.

### **Assessment of Rate of Rooting**

Rooting extension can occur at different speeds and depending on the location and environment, the rate at which that occurs, can determine whether the deeper rooting brings an advantage, disadvantage, or neither, when it comes to better water use and extraction to improve yield. Sinclair, Messina, Beatty, and Samples (2010) wrote that that having roots which grow slowly deeper into wet soil is important to try to maintain the water balance for what is needed in the leaves for transpiration so that severe drought is not met. However, Sinclair et al. (2010) found in a simulation study with soybean that root extension at a rapid rate had no advantage on yield. In fact, in some geographical regions in the US, a greater rate of root extension resulted in

a yield disadvantage in up to 70% of the years (Figure 1.1). This result was found because when roots extend deeper into the soil at a fast rate, they extract more of the soil water, and leave little for later in the growing season when the plants will need water to sustain grain fill (Sinclair et al., 2010). On the other hand, simulations for other areas of the country, such as the Deep South, the faster deeper rooting actually brought an advantage on yield (Sinclair et al., 2010). These results showed that the location and environment plays a key role on whether or not faster or slower deeper rooting brings a yield advantage (Sinclair et al., 2010).

### **Water Conservation for Improved Drought Tolerance**

Another important drought tolerance mechanism for crops, including maize, is the ability to be conservative in their soil water use (Gholipoor et al., 2013). Under drought, the expression of certain traits allow crop plants to limit their water use earlier in the growing season, which then allows them to have access to soil water later in the growing season during reproductive growth (Kholová et al., 2010; Zaman-Allah, Jenkinson, and Vadez, 2011). Developing cultivars that have conservative soil water use earlier in the drying cycle is crucial for improving drought tolerance. Once plants reach the point of drought where their fraction of transpirable soil water (FTSW) has decreased to approximately 0.25-0.40, their transpiration rate decreases linearly (Sadras and Milroy, 1996). Identifying cultivars that decrease their transpiration rate at a higher FTSW threshold (when FTSW is closer to 1.0) is ideal for drought tolerance because these cultivars conserve their soil water use earlier in the drying cycle. In maize, Gholipoor et al. (2013) discovered 12 hybrids that expressed higher FTSW threshold values. These 12 maize hybrids are of value because they are “hypothesized to have the capability to restrict transpiration

water loss at higher soil water contents and conserve water early in the growing season for use later in the growing season” (Gholipour et al., 2013, p. 27).

One example of a trait that allows for soil water conservation earlier in the drying cycle is the limited-transpiration trait. Many maize lines’ transpiration respond to the increased level of vapor pressure deficit (VPD) (Gholipour, Choudhary, Sinclair, Messina, and Cooper, 2013; Yang et al., 2012). A plant that expresses the limited-transpiration trait decreases its transpiration rate under high VPD conditions (Messina et al., 2015). This decrease in transpiration comes as the plants partially close their stomata. This decrease in transpiration may allow for water conservation during the early part of the season, which leaves more water in the soil for the plant to use later under drought conditions, and therefore could improve yield under drought (Messina et al., 2015).

Limited-transpiration trait studies have also been conducted with many soybean genotypes, including PI 416937, a “slow wilting” genotype. Through visual observations in the field, PI 416937 was known to have delayed wilting under drought-stress, compared to other tested soybean lines (Sloane, Patterson, and Carter, 1990). Researchers hypothesized that perhaps PI 416937 possessed delayed wilting because of characteristics of its root system, such as prolific fibrous rooting in the upper levels of the soil (Hudak and Patterson, 1995). King, Purcell, and Brye (2009) showed that this rooting trait did not bring increased access to soil water. After the slow wilting appeared not to be due to rooting characteristics, an alternative hypothesis was examined. Fletcher, Sinclair, and Allen (2007) discovered in the growth chamber that above a VPD of 2.13 kPa, PI 416937 showed a breakpoint in its transpiration rate. PI 416937 also expressed this limited-transpiration trait in the field at approximately 2 kPa (Gilbert, Holbrook, Zwieniecki, Sadok, and Sinclair, 2011). This conservation of water that the limited-

transpiration trait allows for, accomplishes the same goal as Ludlow and Muchow (1990) discussed: decreasing water use early in the growing season so that more water remains for the plant to use during the reproductive period.

### **Climate Change Education**

In order to adapt to climate change, and to decrease of human impact towards increased climate change, younger generations must be educated about climate change and climate science so that they understand the importance of the topic and the severity of its effects. Starting from a young age in the United States public school systems, teaching climate change in primary and secondary schools is not given high priority (Sharma, 2012). In public schools, almost all students learn about climate change to some extent through their middle school science courses and their high school biology course; only approximately 3-4% of students do not receive climate change instruction (Plutzer et al., 2016). A study surveying 1,500 public middle- and high-school teachers found that the average science teacher spends 1-2 hours per year covering climate change in class, specifically discussing recent global warming (Plutzer et al., 2016). Therefore, even though 96-97% of public middle and high school students are learning about climate change, 1-2 hours of instruction does not provide a sufficient allotment of time to deeply study climate change. Because of this, many students are entering university without having had any formal climate change instruction (Kudrna, Shore, and Wassenberg, 2015).

To increase climate change education, schools may increase the number of hours dedicated to climate change in the classroom. However, the problem is more complex due to teachers' preparedness (or lack of) in climate change. Public school teachers are not sufficiently trained to teach science (Aalderen-Smeets, Molen, and Asma, 2011). Of the 1,500 public middle-

and high-school teachers interviewed in a Plutzer et al. study (2016), less than half of the teachers reported that they had any formal climate science instruction in college.

Since many school students in the United States are lacking climate change instruction, student opinions of climate change are commonly formed through influence from media, personal experiences, and the influence of society (Hmielowski, Feldman, Myers, Leiserowitz, and Maibach, 2014; Krosnick and MacInnis, 2010; Lorenzoni and Pidgeon, 2006). Consequently, when people outside the scientific community rely on personal experiences and outside sources, such as media, in attempts to understand climate change, they may be led to bias and incorrect understandings of the causes (Weber and Stern, 2011). When misconceptions of a scientific issue are formed, these misconceptions can prevent people from learning the actual information that is supported through science (Chi, 2005; Vosniadou and Brewer, 1992).

Deitz and Stern (2008) discuss the difficult task to communicate scientific information, such as climate change, to the public in a clear and effective manner, even with information being produced at the fastest rate in history. One reason the task is so difficult may be because of the complexity of comprehending climate change. Climate change is difficult to understand because of several complex factors: the greenhouse gases which cause climate change are invisible; most Americans do not live in an area where they are greatly impacted by climate change; and warning signals of climate change are difficult to see (Moser, 2009; National Research Council, 2009).

As climate change is difficult to communicate, the university seems like a proper setting for it to be taught in detail. While there is a large volume of literature based on school-aged climate change education, very little work has been published on climate change education in the U.S. university system. Chapter 4 of this dissertation will help shrink this gap in the literature.

## Dissertation Overview

Chapters 2 and 3 of this dissertation are plant science research chapters and they focus on improved drought tolerance in maize. The research presented in Chapters 2 and 3 involves two lines of maize (*Zea mays* L.) that have been previously studied by Chimenti, Marcantonio, and Hall (2006) – a high osmotic adjustment line (HOA) and a low osmotic adjustment line (LOA). Chimenti et al. (2006) compared the two lines of maize with diverging levels of OA to understand the value leaf OA has in increasing yield under drought-stress. In their study, Chimenti et al. (2006) saw the LOA Line wilted earlier than the HOA Line. While hypothesizing on what could be the cause of the delayed wilting in the HOA Line, Chimenti et al., (2006) mentioned that perhaps the HOA Line possessed deeper rooting as a result of OA. If the greater leaf OA of the HOA Line extended to greater OA in the root tips, then there was the possibility for greater root extension due to a greater turgor pressure. That is, the roots of the HOA Line would be able to grow deeper into the soil profile, access more soil water, and therefore have delayed and less severe wilting, compared to the LOA Line. In fact, Chimenti et al., (2006) reported that the HOA Line extracted more water than the LOA Line in both of their two field experiments. However, Chimenti et al., (2006) did not examine OP differences in the root tips of the two lines of maize nor did they measure rooting depths of the two lines.

In response to this hypothesis, the specific research objectives of Chapter 2 were to (1) repeat the leaf osmotic measurements as done in Chimenti et al. (2006) for additional documentation of differences in leaf OA, (2) determine whether differences in OA between the two lines occurred in the root tips during drought, and (3) determine whether there were rooting length differences between the HOA Line and the LOA Line.

Chapter 3 of this dissertation examined the differences in water use between the two lines as a possible alternative hypothesis for the delayed wilting in the HOA Line. The specific research objectives of Chapter 3 were to: to (1) determine transpirational differences between the two lines under different VPD conditions, (2) document possible differences between the two lines in the transpiration response curves to soil drying, and (3) determine possible difference between the two lines in their rate of soil water extraction.

Chapter 4 of this dissertation is the educational research chapter that focuses on how university professors teach undergraduates about climate change. The population of this descriptive research study was comprised of NC State faculty who have taught an undergraduate course that contained one of the following phrases in their course description: “climate change,” “global warming,” or “environmental impact.” Twenty-four professors completed a cross-sectional survey questionnaire that sought to understand how they teach about climate change. The specific research objectives of this chapter were to: (1) understand the professors’ views on climate change, (2) explore which climate change-related content they teach in their courses, and (3) understand which teaching methods they use to teach undergraduates about climate change.

## Chapter 1 Figures

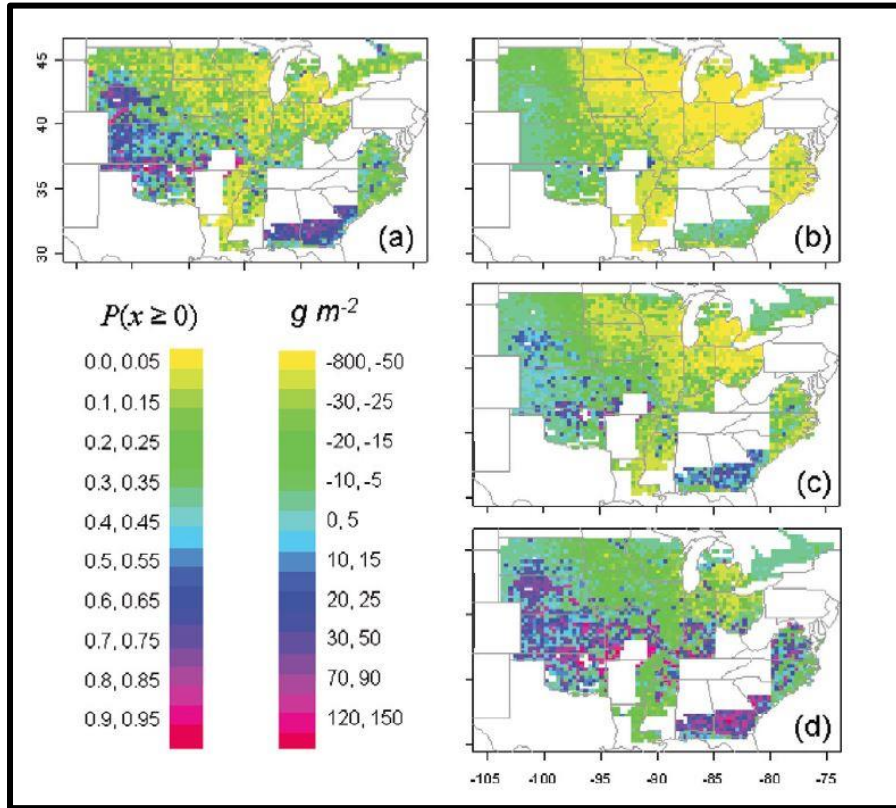


Figure 1.1: “Regional patterns of yield variation due to change in rooting. Probability of yield for simulation with altered trait being greater than the standard simulation (a). Absolute yield difference between the simulation with the altered trait and the standard simulation presented for each grid location for the 75%, median, and 25% quartiles among all years simulated (b to d).” (Sinclair et al., 2010)

## **CHAPTER 2: Comparing Rooting Lengths and Root Osmotic Potential Between Two Lines of Maize**

### **Introduction**

Climate change is one of the largest challenges facing our current and future generations, specifically related to food production (Battisti and Naylor, 2009). One of the major threats of climate change is predicted changes in rainfall patterns. This includes increased droughts in certain areas of the world, both in the number of droughts and the duration of the droughts.

Drought is a major stress that has a negative impact on many plant processes, and therefore often results in crop yield losses (T.R. Sinclair, 2017; Thomas R. Sinclair, Purcell, and Sneller, 2004). A focus must be placed on increasing crop tolerance to drought (Tester, 2011). To lessen the consequences of drought, one physiological mechanism suggested for enhancing crop tolerance is osmotic adjustment (OA) (Blum, 1989; Hare, Cress, and Van Staden, 1998; Moinuddin and Khanna-Chopra, 2004). OA is a plant adaptive response that occurs in response to developing water deficit in the plant at the cellular level (A Blum, 2005). OA is when the osmotic potential (OP) decreases because solutes such as salts, carbohydrates, and amino acids, accumulate in cells (Sanders and Arndt, 2012). Since solutes lower the thermodynamic potential of a solution, OP is expressed as a negative value. The lowering of OP, and consequently lower hydraulic potential in the plant, could be crucial because this increases the hydraulic gradient between plants and soil, and can result in additional water uptake from the soil (Taiz, et al., 2015).

*Zea mays* (L.), commonly known as maize, is an important staple crop grown around the world, at an estimated global production of over one billion metric tons (National Corn Growers

Association, 2018). Since maize is usually grown during the warmer, drier months of the year, maize is susceptible to drought conditions. A study conducted by Chimenti, Marcantonio, and Hall (2006) examined the possibility of selecting maize lines with the OA to increase yield of maize. Under field conditions at Buenos Aries, Argentina, they compared the response to drought of two maize lines, which were selected specifically for differences in leaf OA when subjected to drought. Chimenti et al. (2006) referred to these two lines as a high osmotic adjustment line (HOA) and a low osmotic adjustment line (LOA). They observed that the LOA Line suffered from more severe wilting in a drought treatment than the HOA Line. Under drought conditions, the HOA Line had a significant decrease in leaf OP, while no change in leaf OP was observed in the LOA Line. From their results, Chimenti et al., (2006) concluded that OA in the leaves can add to drought tolerance in maize.

Chimenti et al., (2006) expanded the hypothetical view of the greater drought tolerance of the HOA Line to include deeper rooting by this line as a result of OA. Specifically, if the greater leaf OA of the HOA Line extended to greater OA in the root tips, then there was the possibility for greater root extension due to a greater turgor pressure. That is, the roots of the HOA Line would be able to grow deeper into the soil profile, access more soil water, and therefore have delayed and less severe wilting, compared to the LOA Line. In fact, Chimenti et al., (2006) reported that the HOA Line extracted more water than the LOA Line in both of their two field experiments. However, Chimenti et al., (2006) did not examine OP differences in the root tips of the two lines of maize nor did they measure rooting depths of the two lines.

Consequently, the study of Chimenti et al. (2006) was incomplete in fully resolving the apparent basis of the greater drought tolerance of the HOA Line as compared to the LOA Line. In particular, the objectives of the investigations presented here was to (1) repeat the leaf osmotic

measurements as done in Chimenti et al. (2006) for additional documentation of differences in leaf OA, (2) determine whether differences in OA between the two lines occurred in the root tips during drought, and (3) determine whether there were rooting length differences between the HOA Line and the LOA Line. These objectives were pursued by both studies under stable environmental conditions of plants grown in pots in a growth chamber and in the variable natural environment in the field.

## **Materials and Methods**

### **Plant Material**

The two lines of maize used in Chimenti et al. (2006) were the focus of this research. These two lines were developed by initially screening for OA among 20 elite inbred maize lines that originated from Argentina's flint germplasm adapted to temperate environments. These 20 lines were provided by the Maize Breeding Program of the Pergamino Experiment Station of the National Institute of Agricultural Technology (Chimenti et al., 2006). The screen for OA was done during their early vegetative development (Chimenti, Cantagallo, & Guevara, 1996). The two lines having the most divergent OA, that is, extremely high OA and extremely low OA, were crossed. The F1 seed obtained from the cross were sown and selfed to create a F2 population. The F2 seed were sown and also selfed. The length of the seedling shoot of 141 F3 families were evaluated when subjected to a -0.8 MPa osmotic solution. These results were used to select the 16 F3 families having the lowest and the 16 F3 families having the highest seedling extension. The two sets of families were each bulked forming two breeding populations. The assumption was that the populations formed based on shoot extension reflected differences in OA under

water-deficit conditions. Hence, the selection process of Chimenti et al. (2006) resulted in lines of similar genetic background but with potentially differing levels of OA.

Chimenti et al. (2006) subsequent did field tests using rain shelters in which leaf OA and soil water extraction was measured with developing soil water deficit. The lines were confirmed to have divergent expression of leaf OA, with the one line showing no leaf OA and the second line showing leaf OA up to 0.54 and 0.41 MPa in their two experiments.

## **Plant Growth**

Maize plants were grown in tall pots: 104 cm tall by 10 cm wide, with a volume of 8.4 L. These pots were custom built from polyvinyl chloride (PVC) pipe and a flat PVC cap with a hole for water drainage. A unique feature for these experiments was that a plastic liner made of 6 mil poly tubing (Uline, Pleasant Prairie, WI) was installed in each pot. The use of the poly tubing liner allowed the soil column to be easily removed from the pot for ready examination of root extension with length. Poly tubing was cut to be 120 cm long, which allowed excess tubing to be folded over the top of the 104 cm tall pots. The bottom of the liner was sealed using a tabletop poly bag sealer (Uline, Pleasant Prairie, WI). Holes were then cut into the bottom of each liner to allow for water drainage. One liner was placed into each pot and carefully packed along the pot's edge to decrease the chance of creases, which could alter root extension.

After the liners were inserted in the pots, a loamy sand mineral soil (8.8% clay, 18.2% silt, and 73% sand) was put in the liners. Soil was filled into the pots so they would possess uniform characteristics and volume to maintain a constant bulk density across pots. This procedure involved adding three uniform-sized scoops of soil to the pot, pausing to tap the

bottom of the pots on the floor three times to allow the soil to settle, and then repeating this procedure.

All of the seed used in these experiments were shipped from Buenos Aires, Argentina, and were then stored in a refrigerator in the laboratory at North Carolina State University. Seeds were germinated at 29°C in “ragdolls,” which were wet germination towels rolled together with seed inside. The seeds for germination were chosen to be as uniform as possible in their shape, color, and size. At the start of each experiment, three seedlings were transplanted into each of fifteen pots for each of the two maize lines. After transplanting, plants were given the recommended amount of a nutrient solution on the soil surface (nitrogen 17.9 mM, phosphorus 4.0 mM and potassium 9.0 mM; (MaxiGro, General Hydroponics, Sebastopol, CA). This nutrient solution was also applied to the plants on the soil surface one day before the dry-down treatment started. Approximately 4 to 8 days after transplanting, a uniform population of plants was achieved by thinning each pot to one plant by retaining plants of consistent size across all pots. This thinning of the number of plants in pots typically occurred when the plants were at the V1 stage. All plants were watered as needed.

### **Growth Chamber Experiments**

Three experiments were conducted in a walk-in controlled environment chamber at North Carolina State University’s Phytotron (Chamber A-21). Pots were organized so that two plants of the same line were never sitting side-by-side. Each day pots were moved around the chamber to avoid any location effects based on their position in the chamber. Chamber temperature was set to 26°C during the day and 18°C during the night. The chamber was set to 14 hours of daylight and 10 hours of darkness. Photosynthetically active radiation in the growth chambers was an

average of  $533 \mu\text{mol m}^{-2} \text{s}^{-1}$ . Atmospheric relative humidity in the chamber was controlled by humidifiers and dehumidifiers placed in the chamber. Temperature and relative humidity were logged every five minutes using data loggers (Lascar Electronics, Erie, PA).

Each of the three experiments was done at a different humidity to determine if humidity conditions might influence the development of OA in the plants. The humidity condition with the highest vapor pressure deficit (VPD) was 2.44 to 3.22 kPa (GC-HVPD). The moderate humidity experiment (GC-MVPD) had a VPD range of 1.96 kPa to 2.45 kPa. The lowest VPD experiment (GC-LVPD) ranged between 1.47 to 1.76 kPa (full details are given in Table 2.1). Dehumidifiers and humidifiers were checked frequently to ensure the humidity level was being maintained for the desired VPD conditions.

Once plants reached the V3 stage, the dry-down experiment was initiated. The night before each experiment started, pots were watered until dripping to ensure soil water content was saturated. Pots were slightly elevated off the ground, supported by stacks of flat wooden stakes, so the water could drain out of the drainage hole easier. The pots were left to drip overnight so the soil could drain to pot capacity. In the morning, pre-dawn, the soil was covered with aluminum foil, around the plant stem, to ensure virtually no water loss would occur through soil water evaporation. Pots were then weighed and their initial weights were recorded. Each day immediately after the chamber lights came on, all pots were weighed to allow calculation of daily water loss via transpiration.

Twelve plants of each line were designated to be subjected to the drought-stress treatment (DS) and three plants were designated for the well-watered treatment (WW). For each experiment, plants were assigned to the DS and WW treatments by examining water loss from each plant for the first three days of the experiment. For each of the two lines, one WW plant

was selected with low levels of water loss, one with mid-level water loss, and one with high levels of water loss. This was done to ensure all levels of transpiration would be present in both the DS and WW plants. The remaining twelve pots of each line were subjected to the DS treatment. During the dry-down, WW plants were re-watered daily to maintain a pot weight of 250 g below their initial weight. The DS plants dried as transpiration allowed.

Daily transpiration data were used to calculate the normalized transpiration ratio (NTR) for all DS plants (Serraj, Allen, and Sinclair, 1999). The transpiration rate was calculated by the difference in pot weights from day to day. The daily transpiration of each DS plant was then divided by the mean transpiration rate of the three WW plants within that same genotype on the same day, which gave the transpiration ratio. By calculating the transpiration ratio, daily variation due to possible daily environmental variation in the chamber was minimized. To reduce variation from plant to plant, the daily normalized transpiration rate (NTR) was calculated for each DS plant. NTR was calculated by dividing the plant's normalized transpiration ratio by the plant's average transpiration ratio during the first three days of the dry-down experiment (before the plant started experiencing drought-stress conditions). By definition, at the start of the experiment the values of NTR for each plant were centered on 1.0.

## **Field Experiments**

Two field experiments were conducted in the summer of 2016. The early experiment (FES) was conducted in April and May of 2016 while the late experiment (FLS) was conducted in August and September of 2016 (full details given in Table 2.1). Both field experiments were conducted at the Central Crops Research Station in Clayton, North Carolina (latitude: 35.67, longitude: -78.50) in field number C3B, a 0.4 hectare plot. The entire plot was first sown on 18

April 2016 with Pioneer hybrid P1690. The tall root pots were installed in the soil in two central rows four days after the entire plot was sown. Fifteen pots were installed in each row with 40 cm spacing between each pot. In between the two rows with pots, there were three rows of border maize. The pots were buried into the soil in the field, leaving approximately 10 cm of the pot top above soil level.

Weather data during each experiment was obtained from a weather station on the Central Crop Research Station via the State Climate Office of North Carolina's CRONOS Database. Throughout the FES experiment, the average maximum temperature was 25°C and the average minimum temperature was 19.8°C. The VPD in FES ranged from 0.3 kPa to 2.6 kPa. Throughout FLS, the average maximum temperature was 31.9°C with the average minimum temperature of 26°C. The VPD in FLS ranged from 0.8 kPa to 3.1 kPa.

The soil-drying treatment was imposed in the field by covering each pot to prevent rainfall from entering the pot. The tops of the pot were sealed using aluminum 7 mil 15x18" Dri-Shield™ heavy-duty bags (Uline, Pleasant Prairie, WI). The bag was sealed around the stem of the plant by layering bags and foil at different angles and then sealing with duct tape. Also, the bag was sealed around the top of the pot using duct tape. This pot cover was checked daily to ensure all pots were still covered sufficiently so no rainfall could get into the pots. Since all pots were covered in this manner, the WW plants required daily watering by lifting the foil cover around the pots. The pots were not weighed during the field experiments (due to logistical problems in dealing with the buried pots).

The second field experiment (FLS) was located in the same spot of field as the FES experiment. By the time seed for FLS were sown, the border maize had grown tall and would have shaded new plants sown into pots. Therefore, the border maize in the rows directly

surrounding the two experimentally rows, and between pots within the two experimental rows, were removed. Otherwise the FLS experiment was handled identically as in FES.

### **Leaf Osmotic Potential**

During each growth chamber experiment, leaf “hole punch” samples were taken periodically to compare the leaf OP between the HOA Line and the LOA Line. The leaf punches were taken from a top leaf that did not yet have an exposed collar. Leaf punches (6.4 mm in diameter) were taken from both sides of the mid-vein, approximately half way up the leaf using a paper hole punch made of metal. If the plant was to be harvested on that day, a total of six leaf punches were taken, since leaf damage did not matter. If the plant was not being harvested on that day, only 1 leaf punch was taken, to minimize leaf damage.

Immediately after obtaining the leaf samples, they were placed in plastic bags in deionized water and stored in a refrigerator at 4°C for four hours to obtain rehydrated samples for OP measurement (Arndt, Irawan, and Sanders, 2015). Following the four hour incubation, the samples were lightly tapped dry, wrapped into heavy-duty aluminum foil, sealed into a small plastic bag, and frozen at -80°C.

When OP was to be measured, leaf punches were thawed. In the case where multiple punches were obtained, a single leaf punch was selected for analysis. A syringe needle was used to punch holes in the sample. The sample was then placed into an osmometer (Model 5500, Wescor Inc., Logan, UT) and OP was determined. The sample was measured in the osmometer until two consecutive readings were stable. This procedure usually took approximately 4 to 8 minutes. For each sample, osmotic values (MPa) were obtained by inserting the mean osmolality into the van't Hoff equation as follows:  $\Psi_{\pi} \approx -nRT/V$ , where n is osmolality, R is a gas constant

( $8.32 \times 10^{-6} \text{ MPa m}^3 \text{ mol}^{-1} \text{ K}^{-1}$ ), T is temperature in degrees Kelvin, and V is the volume of the cell in cubic meters.

Leaf OP was graphed against NTR to determine how leaf OP might change as NTR values decrease throughout the soil dry-down cycle. Linear regression analysis was conducted for each maize line. Excel's Data Analysis tool kit was used to calculate the p-values associated with the significance of the regression slopes by using analysis of variance (ANOVA). A two-tailed T-Test was conducted to find the differences between the mean leaf OP values between the two lines.

### **Root Length and Osmotic Potential**

Drought-stressed plants were periodically harvested from both chamber and field experiments by sliding the plastic liner from the pot (containing the plant, roots, and soil) and laid gently on a tabletop. The plastic liner was carefully removed from the soil by using a razor blade to cut the plastic while avoiding cutting of roots. After the plastic liner was removed, the soil was carefully removed from all roots. During the earlier harvest dates, this was a much easier task because the soil was still damp. In the later harvest dates, the soil was much drier and firm, making soil removal from the roots much more difficult. A screwdriver and paintbrush were used to assist with soil removal. Once all soil was removed from the roots, each root was cut from the base of the maize stem and laid on a flat table. Roots were separated into nodal roots and seminal roots and their length measured.

In GC-MVPD and GC-HVPD, there were six harvest dates. The first harvest occurred one day after the dry-down treatment started and followed by harvests occurring every 3 to 4 days. On each harvest day, two DS plants per line were harvested; these plants were chosen

randomly for harvest (see Figures 3.3 and 3.4 for harvest dates). For the GC-LVPD experiment, there were nine harvest days total. The harvests in this experiment were based on plant response to drying. The first harvest day was on the day that the DS plants became noticeably wilted. Three DS plants per line were harvested on this day: two plants per line with more advanced wilting and one that only moderately wilted. The second harvest of three DS plants for each line was based on NTR values: one from each line with the highest NTR values (HOA at NTR=0.75 and LOA at NTR=0.38), one from each line with a medium NTR value (HOA at NTR=0.53; LOA at NTR=0.22), and one from each line with a smallest NTR value (HOA at NTR=0.24; LOA at 0.17). For harvests 3 through 9, DS plants were harvested individually as each plant reached an NTR value of  $\leq 0.1$  (total of 12 DS plants). At the final harvest of each experiment, all WW plants were also harvested to examine their rooting length (total of 6 WW plants in each experiment).

For both field studies, FES and FLS, there were two successful harvest dates per experiment, which increased the number of replicate DS plants to four at each sampling (see Table 2.2 for harvest dates). The first harvest occurred when the DS plants first started to wilt and the second harvest occurred when the DS plants were severely wilted. Initial wilting included first signs of leaf folding and rolling. Severe wilting included severe wilting of leaves throughout the canopy and the loss of visual leaf turgidity. In both field experiments, a third harvest was planned but was not carried out since the plant roots had already reached the bottom of the pots before this harvest was to be done. In both field experiments there were 3 WW plants per line (6 WW plants total), which were harvested during the second harvest.

During each harvest of all experiments, as soon as root length was measured, root tip samples were taken from the five longest nodal roots and the five longest seminal roots (when

possible). The terminal 6 to 6.5 cm of these roots were removed with a clean cut using a razor blade. The root tip samples were placed in small plastic bags with deionized water (nodal roots in one bag, seminal roots in another). The samples were placed in a refrigerator at 4°C for four hours to allow tissue rehydration. Following the rehydration period, the samples were removed from the plastic bag, lightly tapped dry, wrapped into heavy-duty aluminum foil, sealed into a small plastic bag, and frozen at -80°C.

At the time of measuring OP, the root tips were taken out of the freezer and thawed. Plant sap was extracted separately from each root tip by the mechanical pressure of squeezing the tissue sample through a 8 mL 30 gauge insulin syringe. Prior to inserting the roots in the syringes, the syringe tips had been cut off so that sap more readily flowed from the syringe. The sap from each root was excised into a petri dish from which the sample for OP was collected. A pipette was used to retrieve 20 µl of sap and discharge the sap on a round paper sample disc (NC9284913, Wescor Inc., Logan, UT). The sample disc was placed in an osmometer (Model 5500, Wescor Inc., Logan, UT) to measure OP. The sample was analyzed in the osmometer until two consecutive values showed stability in the output, which usually took 4 to 8 minutes. Osmotic values were obtained as described previously for leaves.

Root tip OP was graphed against NTR for the chamber experiments to examine how root tip OP might have changed (for nodal and seminal root tips) as the NTR decreased during soil dry-down. Linear regression analysis was conducted for each maize line. Excel's Data Analysis tool kit was used to calculate the p-values associated with the significance of slopes by using analysis of variance (ANOVA). A two-tailed T-Test was conducted to find the differences between the mean root tip OP values between the two lines.

## Results

### Leaf Osmotic Adjustment

Osmotic adjustment in the leaves was observed by measuring OP of the leaves harvested from plants at various stages in the soil drying cycle. In the GC-LVPD experiment, the leaf OP was measured on 12 leaves of each line. Linear regression of leaf OP and NTR failed to show a significant slope (Figure 2.1) in either line (p-value = 0.11 for the HOA Line; p-value = 0.45 for the LOA Line). These results showed there was no change in OP leaves with increasing water deficit, that is, no osmotic adjustment. However, mean OP across all NTR of the HOA Line and LOA Line differed (p-value = 0.08\*). The mean OP values were -1.45 MPa for the HOA Line and -1.22 MPa for the LOA Line. Therefore, the difference in OP between the two lines was 0.23 MPa.

In GC-MVPD and GC-HVPD, where VPD conditions were higher, significant differences in leaf OP between the HOA Line and the LOA Line were not observed. In GC-MVPD, the mean OP values were -1.16 MPa for the HOA Line, and -1.19 MPa for the LOA Line (p-value=0.84). In GC-HVPD, the mean OP values were -1.12 MPa for the HOA Line, and -1.16 MPa for the LOA Line (p-value=0.76).

### Root Tip Osmotic Potential and Adjustment

Figure 2.2 shows the OP values measured in the GC-LVPD experiment for nodal root tips. Each data point in Figure 2.2 represents the average OP of nodal root tips from the five longest nodal roots from one plant on its harvest day and the associated plant NTR value on the day of harvest. The linear regression of nodal root tip osmotic potential vs. NTR for each line, which is shown in Figure 2.2, presented significant slopes for each line based on ANOVA

analysis (p-value of the HOA Line=0.01\*\* and p-value of the LOA Line=0.08\*). These results show there was a decrease in root tip OP with increasing water deficit, and therefore, osmotic adjustment occurred in each line. Across all NTR levels, the HOA Line and the LOA Line did not differ in their mean OP values, based on the p-value from the two-tailed T-Test (p-value=0.87). The mean OP value for the HOA Line was -1.00 MPa and the mean OP value for the LOA Line was -0.99 MPa.

In the GC-LVPD experiment, OP of the seminal root tips for the HOA and LOA Lines was also analyzed. In the earlier part of the experiment (at higher NTR values), it was difficult to retrieve seminal root data because there were fewer seminal roots than nodal roots, and the seminal roots that were present, were fragile and failed to remain intact. As the soil became drier and the NTR decreased, it was easier to obtain seminal root tip samples because the seminal roots remained intact. Due to the lack of samples from the higher NTR values, only results at NTR values of 0.2 and below were compared between the two lines. In this range, nine samples total were analyzed (5 LOA Line, 4 HOA Line). The two-tailed T-Test did not show significant differences between the HOA and LOA Lines in their seminal root tip OP (p-value=0.40). Although root tip samples were taken during all experiments, root tips from FES and FLS experiments were not analyzed due to the higher VPD conditions of these two field experiments.

### **Root Length**

In both the GC-MVPD (Figure 2.3) and the GC-HVPD (Figure 2.4) experiments, there were no consistent significant differences in rooting length between the HOA Line and the LOA Line during any of the harvest days. Two exceptions arose in the GC-MVPD and GC-HVPD experiments as the HOA Line had significantly longer nodal rooting than the LOA Line during

one of the first two harvests, and during the last harvest (Figures 2.3 and 2.4). In the GC-LVPD experiment, again there were no consistent significant differences in rooting length between the two lines. In Harvest 8 another exception appeared as the LOA Line had significantly deeper nodal rooting than the HOA Line (Figure 2.5). For experiments GC-MVPD, GC-HVPD, and GC-LVPD, no seminal root lengths were analyzed due to the difficulty of collecting seminal roots, as discussed in the root tips OP results section.

In the FES experiment, there were also no consistent significant differences in rooting length between the HOA Line and the LOA Line (Figure 2.6). During the second harvest, the LOA Line had significantly deeper nodal rooting than the HOA Line, but this was not the case during the first harvest (Figure 2.6). In both field experiments, seminal roots were harvested without problems. In the FES experiment, there were no significant differences between the HOA Line and the LOA Line in seminal rooting length (Figure 2.6). As for the FLS experiment, no significant differences were found between the HOA Line and the LOA Line in nodal rooting length (Figure 2.7). In the FLS experiment seminal rooting length measurements, there were also no consistent significant differences in rooting length. However, the LOA Line had significantly deeper nodal rooting than the HOA Line during the first harvest (Figure 2.7).

## **Discussion**

While seeking to understand the value of OA for crop yield under drought-stress, Chimenti et al. (2006) observed that the HOA Line had more OA occurring in its leaves, compared to the LOA Line. The first objective of this research was to repeat leaf osmotic measurements as done in Chimenti et al. (2006). Leaf OP was measured in three growth chamber experiments: GC-MVPD, GC-HVPD, and GC-LVPD.

In the GC-LVPD experiment, where VPD conditions were lower, the HOA Line possessed significantly lower (more negative) leaf OP than the LOA Line ( $p$ -value=0.08\*). The HOA Line had a mean OP value of -1.45 MPa and the LOA Line had a mean OP value of -1.22 MPa resulting in a difference in means of 0.23 MPa. These OP results are similar to the results that Chimenti et al. (2006) measured. In the first experiment of Chimenti et al. (2006), the HOA Line had a mean OP value of -1.15 MPa and the LOA Line had a mean OP value of -0.83 MPa. The difference of these means from their first experiment was 0.27 MPa, which was calculated from Table 1 in Chimenti et al. (2006) by taking the average leaf OP of the water deficit plants across all measurement dates. Similarly, in their second experiment, the HOA Line had a mean OP value of -1.17 MPa and the LOA Line had a mean OP value of -0.95 MPa. The difference of these means was 0.22, which was again calculated from Table 1 in Chimenti et al. (2006). In GC-LVPD, as well as in Chimenti et al. (2006), the HOA Line expressed a more negative OP compared to the LOA Line.

The GC-LVPD experiment and the experiments of Chimenti et al. (2006) also had another factor in common: their VPD conditions. Although the VPD conditions for the experiments in Chimenti et al. (2006) were not reported, nor the timing of the experiments, it seems likely experiments were done under modest VPD conditions. In this area of Buenos Aires later in the season, the temperatures may be cooler (reported 27.0°C and 26.5°C), and considering this cool environment, a lower VPD could be assumed. In contrast to the observations in low VPD experiments, the OP results from the GC-MVPD and GC-HVPD experiments in which VPD conditions were higher, the HOA Line did not possess lower (more negative) leaf OP than the LOA Line. In GC-MVPD, the HOA Line had a mean OP value of -1.16 MPa and the LOA Line had a mean OP value of -1.19 MPa. The difference of these means

was not significant ( $p$ -value=0.843). In GC-HVPD, the HOA Line had a mean OP value of -1.12 MPa and the LOA Line had a mean OP value of -1.16 MPa. The difference of these means was also not significant ( $p$ -value =0.76). The basis for VPD being a factor in the expression of OP between the two maize lines is not resolved in these experiments.

Related to OP, the GC-VLPD and the experiments of Chimenti et al. (2006) had one difference. In the experiments of Chimenti et al. (2006), the OP values become more negative as the drought worsens. In the GC-VLPD experiment (Figure 2.1) as the dry-down becomes more severe, the OP values of each line increased, instead of becoming more negative as would be expected as a result of OA under increasing water deficit. However, the data from GC-LVPD appear quite scattered, with very low  $R^2$  values (0.11 for the HOA Line; 0.08 for the LOA Line). More leaf OP samples need to be collected and analyzed to confirm the lack of OA in either line in the GC-LVPD experiment.

As for OA, Chimenti et al. (2006) witnessed leaf OA occurring as drought became more severe. Chimenti et al. (2006) discovered, as the name indicated, that the HOA Line had much more leaf OA than the LOA Line as calculated in the difference in OP on each day between WW and DS plants. In their first experiment, at 35 days after sowing (DAS) the leaf OA of the HOA Line was 0.01 MPa. It increased to 0.54 MPa by day 57 DAS. The leaf OA of the LOA Line was also at 0.01 MPa at 35 DAS but it only climbed to 0.06 MPa at 57 DAS. In the second experiment of Chimenti et al. (2006), the same trend occurred. At 50 DAS, the HOA Line had 0.02 MPa leaf OA and it increased to 0.41 MPa by 70 DAS. The LOA Line had lower leaf OA with 0.03 MPa at 50 DAS and 0.07MPa at 70 DAS. The basis for the discrepancy between the OA observations for the HOA line in Chimenti et al. (2006) experiments and the GC-LVD is unknown.

The two chamber experiments in higher VPD conditions also failed to show OA occurred in the leaves. None of the slopes between leaf OP and NTR were significantly different from zero, signifying no OA occurred (GC-MVPD: p-value of HOA Line=0.25, p-value of LOA Line=0.43; GC-HVPD: p-value of HOA Line=0.57, p-value of LOA Line=0.30).

The second objective, and a primary focus of this current research, was to determine whether possible differences in expression of leaf OP and OA extended to the root tips during drought. As the HOA Line wilted later than the LOA Line in Chimenti et al. (2006), one of the hypotheses to explain delayed wilting was a longer root system, which might be the product of higher OP in the root tips of the HOA Line. In the GC-LVPD experiment, nodal root tip OP was compared between the two lines and it was found that the mean nodal root tip OP values were not significantly different between the LOA and HOA Lines (p-value=0.87). In the GC-VLPD experiment, OP values of the seminal root tips with different NTR values were also measured and no significant differences between the LOA and HOA Lines were found. Hence, the results from the two root types do not support the hypothesis of lower OP in the roots of HOA Line

Root tip OA was also examined in the GC-LVPD experiment by plotting OP against NTR throughout the soil drying cycle on different root harvest days. Figure 2.2 shows the linear regression of nodal root tip osmotic potential vs. NTR for each line, which presented significant slopes based on ANOVA analysis (p-value of the HOA Line=0.01\*\* and p-value of the LOA Line=0.08\*). These results show that within each line there was a decrease in root tip OP with increasing water deficit indicating that OA occurred in both the HOA Line and the LOA Line. Hence, the hypothesis of Chimenti et al. (2006) that OA adjustment in the root tip occurred with soil water deficit, but importantly the two lines did not differ in this characteristic.

Although no significant differences were found between the two lines in root tip OP, measuring rooting length was a priority of this research. The HOA Line could have possessed deeper rooting for another reason besides increased pressure due to root tip OP, and could have served as the reason for delayed wilting. The third objective of this research was to determine whether there were rooting length differences between the HOA Line and the LOA Line. Rooting length was measured in the three growth chamber experiments and two field experiments. Overall, from these five independent experiments, no rooting length differences were observed between the HOA Line and the LOA Line, in neither nodal roots nor seminal roots (Figures 2.3, 2.4, 2.5, 2.6, and 2.7). Root development was tracked with sampling at different times through all five experiments, but no significant differences were shown consistently in any stage of growth.

The results of the root experiments presented here leave open the question of why the HOA Line possessed delayed and less severe wilting compared to the LOA Line in Chimenti et al. (2006). If differences in OP in the root tips, nor differences in rooting length, can explain observed differences in wilting between the two lines under drought, perhaps an alternative hypothesis related to difference in the water use by the two lines through time might explain the observations of Chimenti et al. (2006). Differences in the timing and amount of soil water extraction could lead to differing responses to progressive soil drying as done by Chimenti et al. (2006) and done here in both growth chamber and field experiments. In the next chapter of this dissertation, the temporal dynamics of water use and transpiration will be examined to gain further understanding whether the HOA Line and the LOA line may differ in their water use and how that would influence their response to developing drought.



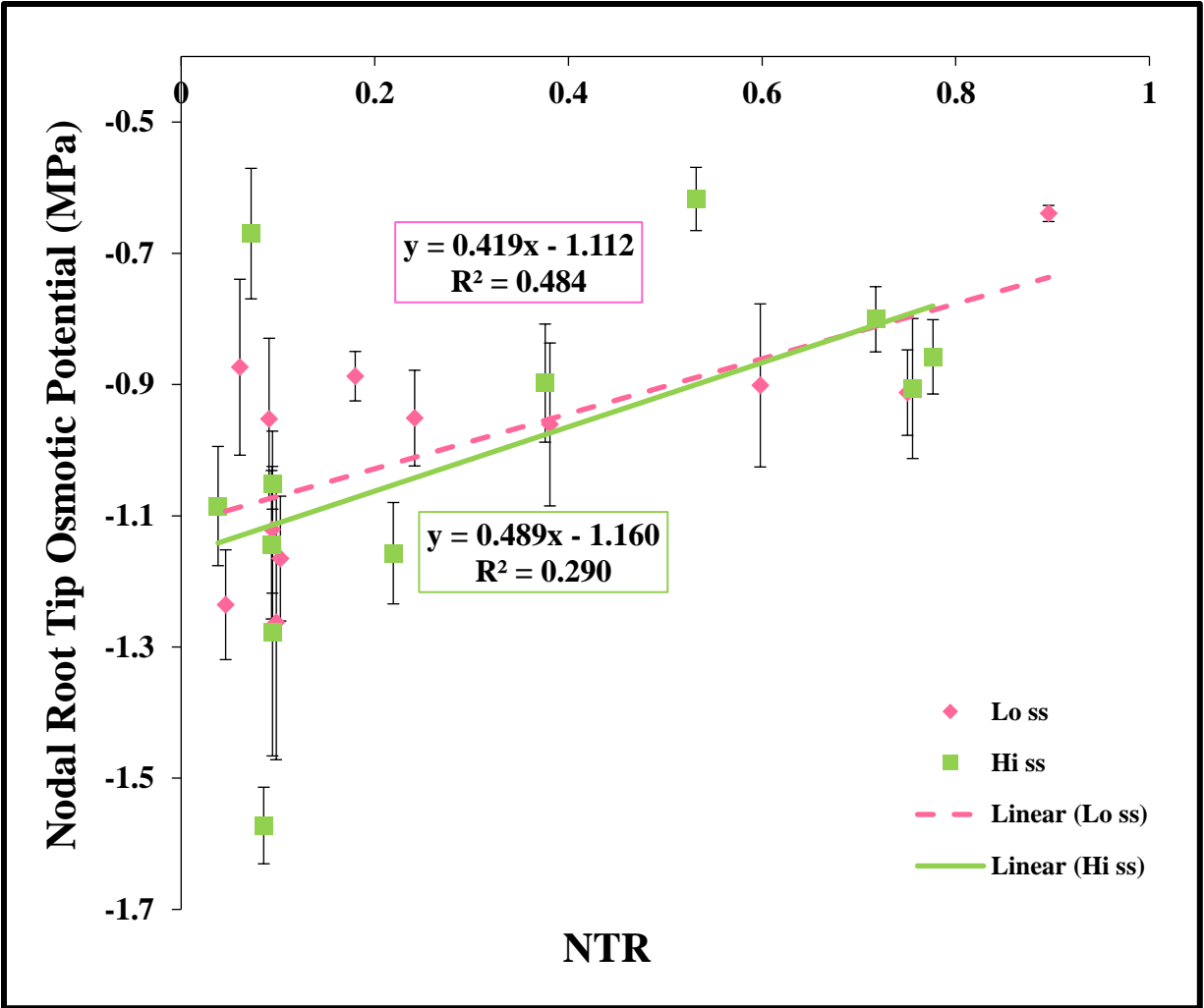


Figure 2.2: GC-LVPD: Nodal Root Tip Osmotic Potential vs. NTR. The regression between NTR and osmotic potential of nodal root tips. Each data point represents the average OP of nodal root tips from the five longest nodal roots from one plant on its harvest day. The NTR values were calculated on the day of harvest. The HOA Line is represented with green data points and the LOA Line with pink. The error bars represent standard error. The slope of the HOA Line was significant (p-value of 0.08\*). The slope of the LOA Line was significant (P-Value of 0.01\*\*). The LOA Line OP values are not significantly different from the HOA Line OP values (P-Value of 0.87 from the T-Test).

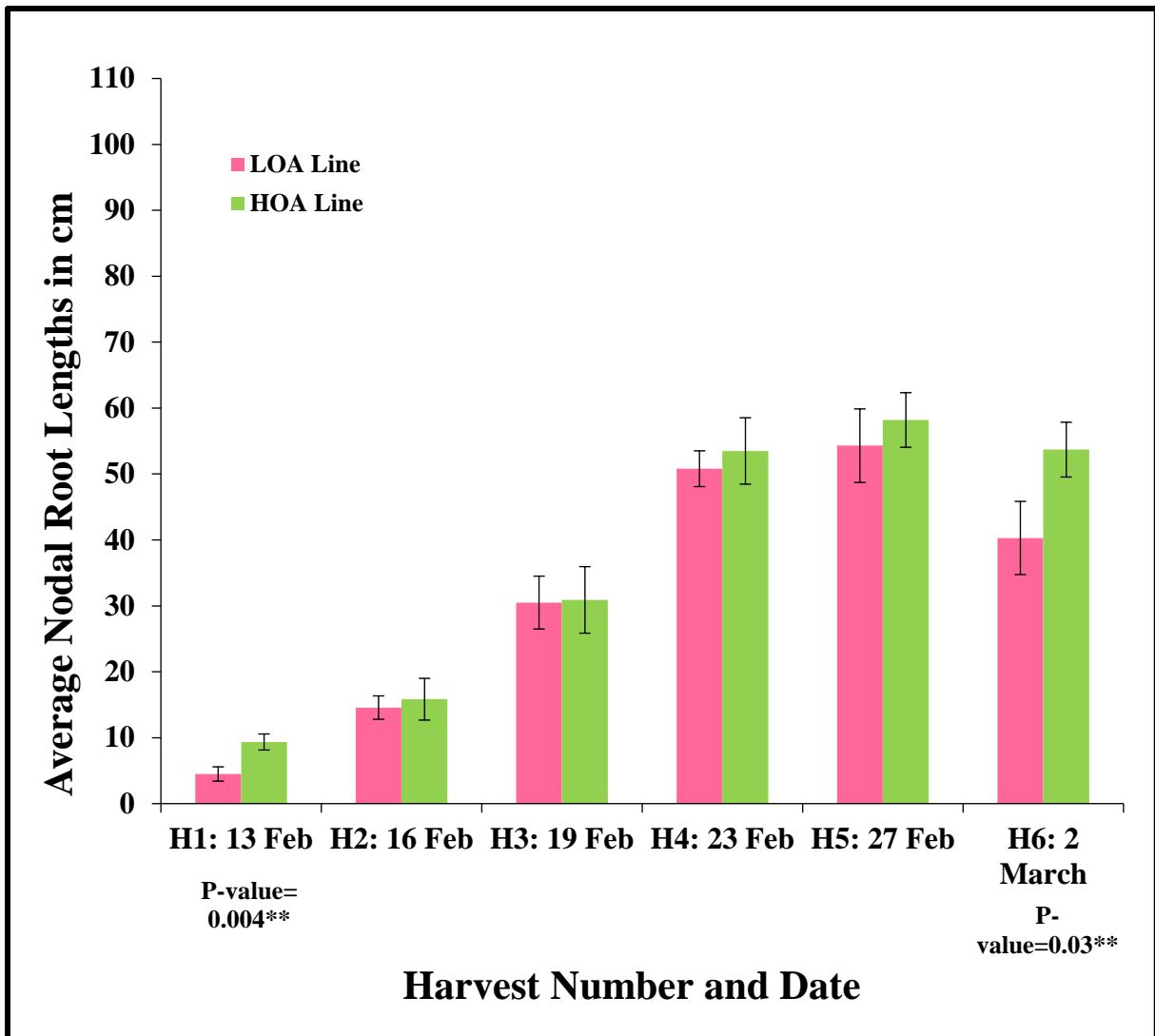


Figure 2.3: GC-MVPD: Comparing the Average Nodal Root Lengths of the HOA and LOA Lines for Each Harvest Day. This figure allows for a comparison of nodal root length between the HOA Line and the LOA Line. Average nodal roots lengths (cm) per line for each harvest day were shown from the GC-MVPD experiment. The error bars represent the standard error within each line of maize for each harvest day. The green bars represent the HOA Line and the pink bars represent the LOA Line. One-tailed T-test was conducted from each harvest day to see if there were significant differences in rooting length between the HOA Line and the LOA Line. Significance of  $p\text{-value} \leq 0.05^{**}$  appeared during Harvest 1 and Harvest 6.

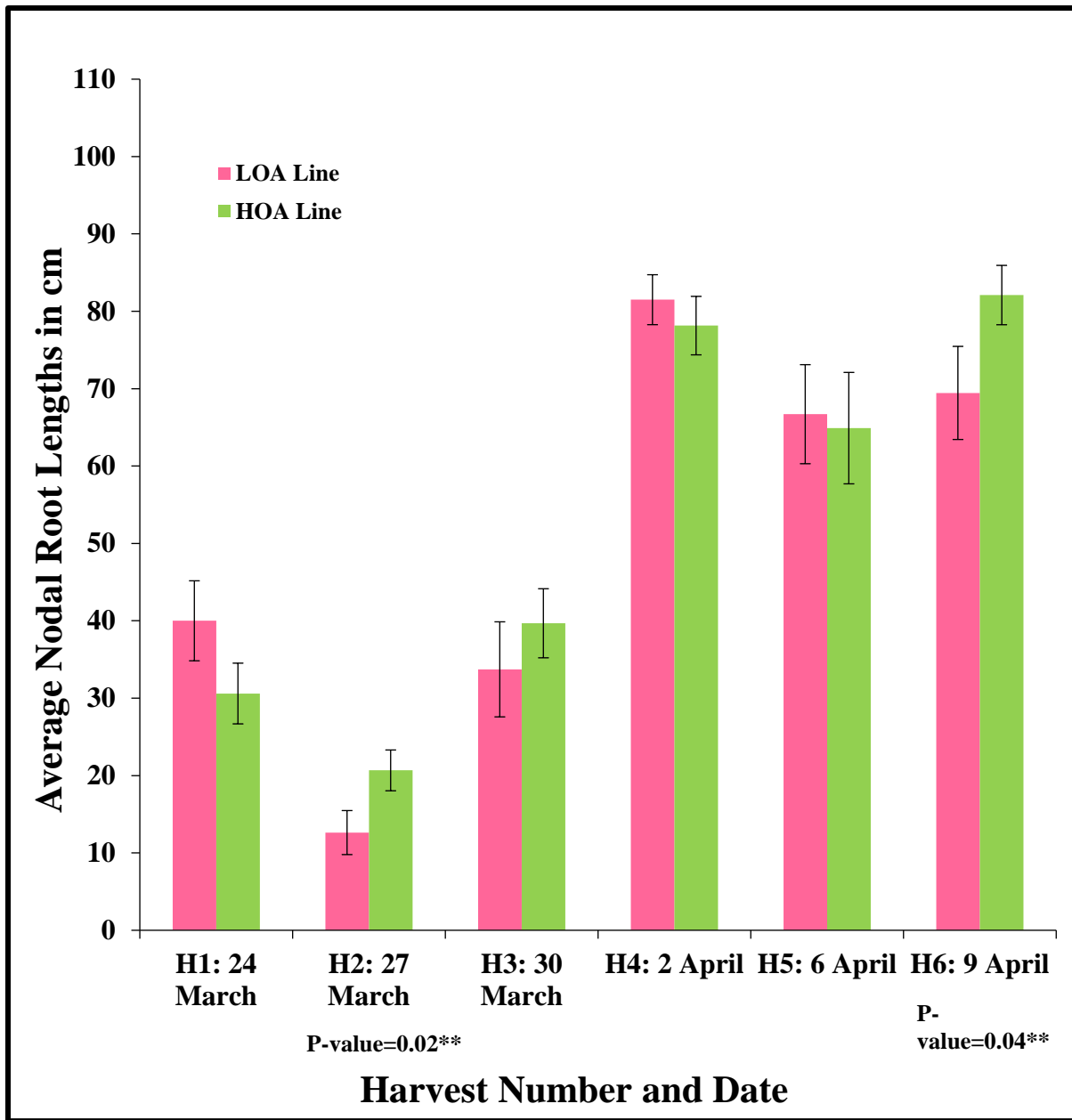


Figure 2.4: GC-HVPD: Comparing the Average Nodal Root Lengths of the HOA and LOA Lines for Each Harvest. This figure allows for a comparison of nodal root length between the HOA Line and the LOA Line. Average nodal roots lengths (cm) per line for each harvest day were shown from the GC-HVPD experiment. The error bars represent the standard error within each line of maize for each harvest day. The green bars represent the HOA Line and the pink bars represent the LOA Line. One-tailed T-test was conducted from each harvest day to see if there were significant differences in rooting length between the HOA Line and the LOA Line. Significance of  $p\text{-value} \leq 0.05^{**}$  appeared during Harvest 2 and Harvest 6.

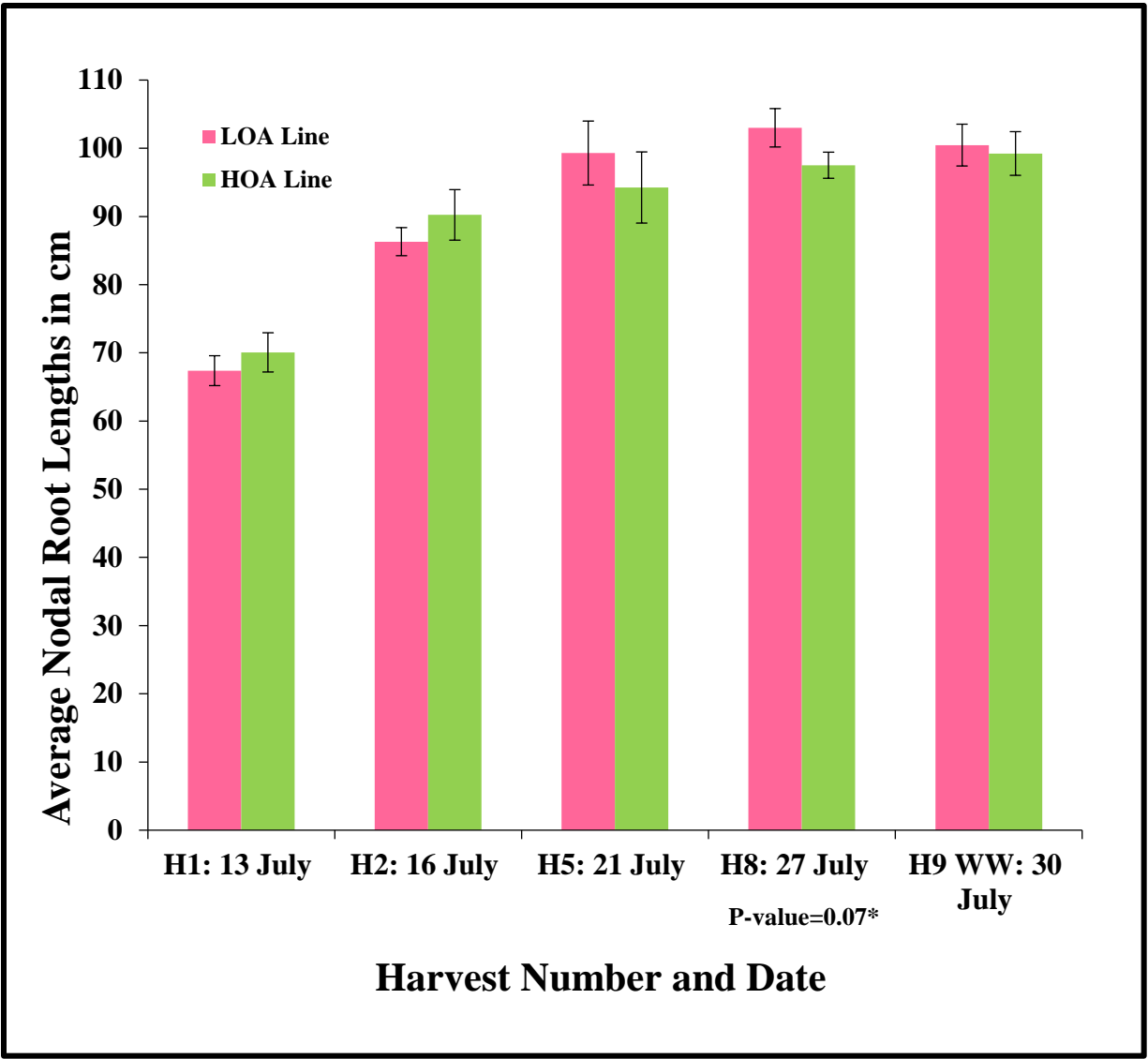


Figure 2.5: GC-LVPD: Comparing the Average Nodal Root Lengths of the HOA and LOA Lines for Each Harvest. This figure allows for a comparison of nodal root length between the HOA Line and the LOA Line for the GC-LVPD experiment. Average nodal roots lengths (cm) per line were shown only for the harvests that contained plants from both the HOA Line and the LOA Line (during Harvests 3, 4, 6, 7, and 9 only drought-stressed plants from only the HOA Line or the LOA Line – not from both – were harvested). Harvest 9 shown above only contained WW plants. The error bars represent the standard error within each line of maize for each harvest day. The green bars represent the HOA Line and the pink bars represent the LOA Line. One-tailed T-test was conducted from each harvest day to see if there were significant differences in rooting length between the HOA Line and the LOA Line. Significance of  $p\text{-value} \leq 0.10^*$  appeared during Harvest 8.

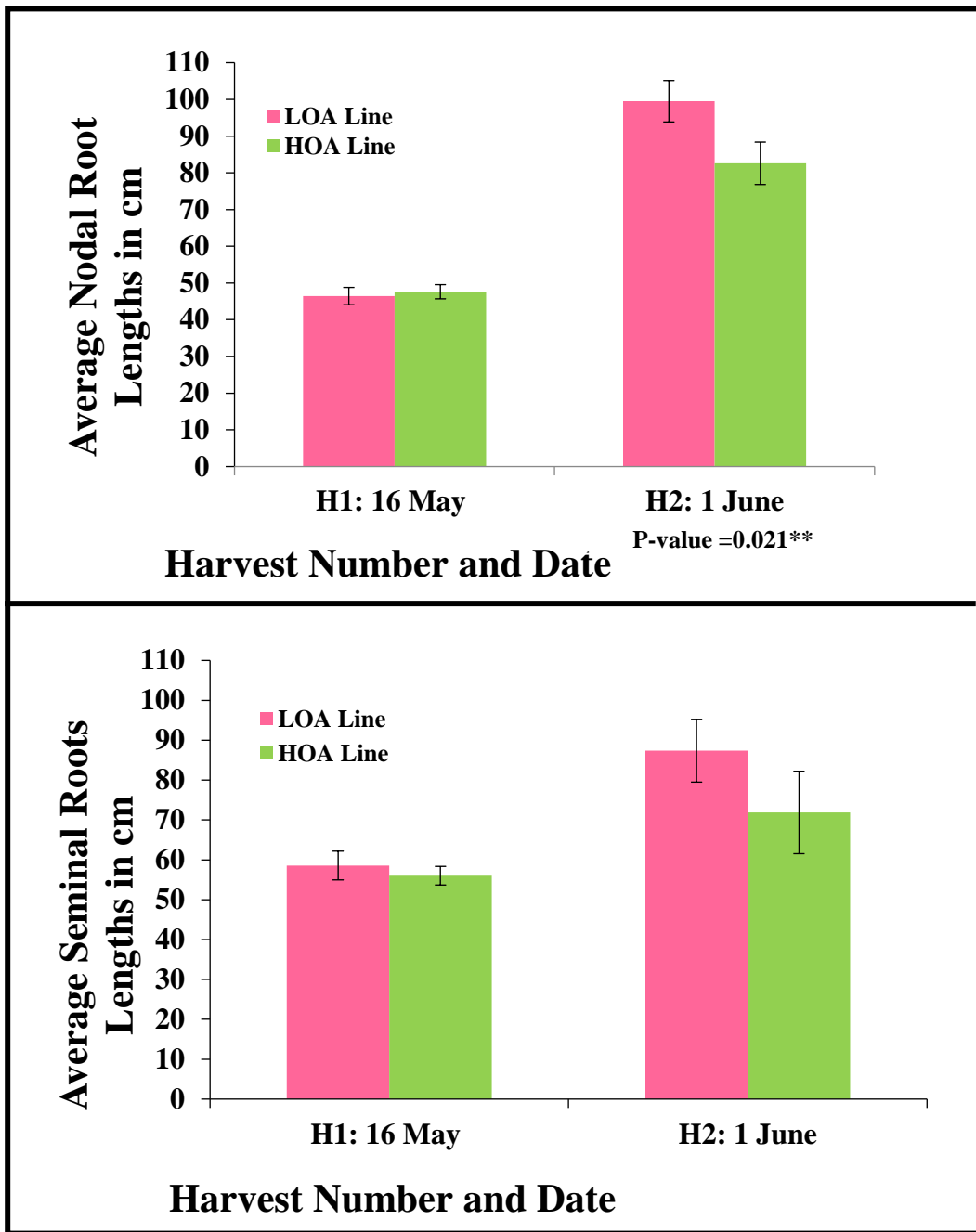


Figure 2.6: FES: Comparing the Average Root Lengths of the HOA and LOA Lines for Each Harvest (top panel, nodal; bottom panel, seminal). Figures above allow for a comparison of nodal (top panel) or the seminal (bottom panel) root length (cm) between the HOA Line and the LOA Line during the FES experiment. The error bars represent the standard error within each line of maize for each harvest day. The green bars represent the HOA Line and the pink bars represent the LOA Line. One-tailed T-test was conducted from each harvest day to see if there were significant differences in rooting length between the HOA Line and the LOA Line. Significance of  $p\text{-value} \leq 0.05^{**}$  appeared only in nodal roots during Harvest 2.

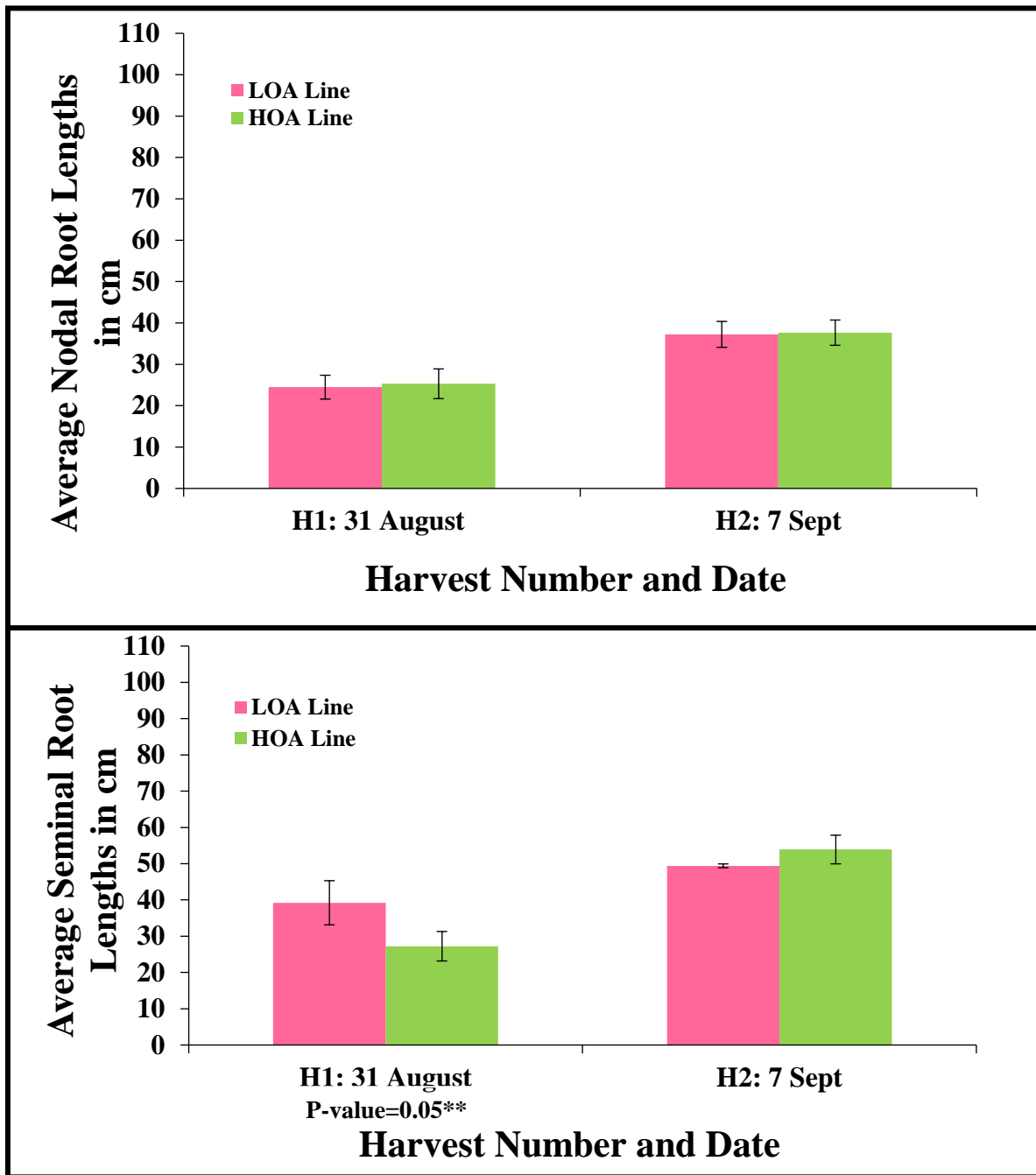


Figure 2.7: FLS: Comparing the Average Root Lengths of the HOA and LOA Lines for Each Harvest (top panel, nodal; bottom panel, seminal). Figures above allow for a comparison of nodal (top panel) or the seminal (bottom panel) root length (cm) between the two lines during the FLS experiment. The error bars represent the standard error within each line of maize for each harvest day. The green bars represent the HOA Line and the pink bars represent the LOA Line. One-tailed T-test was conducted from each harvest day to see if there were significant differences in rooting length between the HOA Line and the LOA Line. Significance of p-value  $\leq 0.10^*$  appeared only in seminal roots during Harvest 1.

## Chapter 2 Tables

Table 2.1: Overview of each experiment with their environmental conditions and important dates.

Experiment Name	Growth Chamber Mid VPD	Growth Chamber High VPD	Growth Chamber Low VPD	Field Early Summer	Field Late Summer
Name Abbreviation	GC-MVPD	GC-HVPD	GC-LVPD	FES	FLS
Classification	Maize Rooting Length Experiments				
Experiment Location	NCSU Phytotron Growth Chamber	NCSU Phytotron Growth Chamber	NCSU Phytotron Growth Chamber	Central Crops Res. Station Field	Central Crops Res. Station Field
VPD Level	1.96kPa – 2.45kPa	2.44kPa – 3.22kPa	1.47kPa – 1.76kPa	0.3kPa – 2.6kPa	0.8kPa – 3.1kPa
Temperature	26°C Day/18°C Night	26°C Day/18°C Night	26°C Day/18°C Night	25°C–19.8°C Average Daily Max & Min	31.9°C-26°C Average Daily Max & Min
Light Hours	14 Hr Day/10 Hr Night	14 Hr Day/10 Hr Night	14 Hr Day/10 Hr Night	Field Conditions	Field Conditions
Ragdolls Started	27 Jan 2015	2 March 2015	12 June 2015	19 April 2016	15 Aug 2016
Seeds Transplanted	30 Jan 2015	6 March 2015	15 June 2015	22 April 2016	18 Aug 2016
Plants Thinned	4 Feb 2015	17 March 2015	23 June 2015	1 May 2016	21 Aug 2016
Start of Dry-Down	12 Feb 2015	23 March 2015	30 June 2015	10 May 2016	29 Aug 2015
Last Root Harvest	2 March 2015	9 April 2015	30 July 2015	1 June 2016	7 Sept 2016
Number of Harvests	6	6	9	2	2
Criteria of When to Harvest	Designed around the number of days: first day after V3; then every 3-4 days	Designed around the number of days: first day after V3; then every 3-4 days	First harvest took place when wilting was noticeable; then by NTR levels	First harvest when plants first started to wilt; second harvest at severe wilting	First harvest when plants first started to wilt; second harvest at severe wilting

## CHAPTER 3: Dynamics of Temporal Water Use in Two Lines of Maize

### Introduction

Drought is the major challenge for increasing crop yields in many parts of the world (T.R. Sinclair, 2017). As drought worsens with climate change, improved crop drought tolerance is crucial for our existence. Studies to understand the response of plant gas exchange in response to soil drying have been done with several crop species, including maize (*Zea mays* L.) (Gholipour et al., 2013). One of the methods used to examine this type of data is to use a two-segment linear regression of normalized transpiration rate (NTR) versus fraction of transpirable soil water (FTSW). At high soil water content, i.e. high FTSW, transpiration rate remains essentially constant as the soil dries. Ultimately, the soil dries to a point at which there is a linear decrease in transpiration with continued soil drying. As an example of a study that examined NTR vs. FTSW was Gholipour et al. (2013). In their study, 24 maize hybrids were tested and the FTSW value at the breakpoint between the two segments among those hybrids varied significantly from 0.33 to 0.51 (Gholipour et al., 2013). The genotypes that had a higher FTSW threshold, meaning they decreased their transpiration rates earlier as the soil dried, were more conservative in their soil water use and may be better suited for arid regions. Consequently, genotypes with a higher FTSW threshold may be more drought tolerant.

As mentioned in Chapter 2 of this dissertation, many studies have explored the possibility that a useful mechanism for improved drought tolerance in crops is osmotic adjustment (OA) (Blum, 1989; Blum, 2017; Hare et al., 1998; Moinuddin and Khanna-Chopra, 2004). This approach is based on an accumulation of solutes in the cells under increasing water deficit that result in a decrease (more negative) of tissue osmotic potential. One example of an apparent

benefit of OA was in maize in the study of Chimenti, Marcantonio, and Hall (2006) which compared two lines of maize with diverging levels of leaf OA. They concluded that OA can lead to improved drought tolerance in maize under water deficit before and during flowering and that under well-watered conditions, the OA trait brings no yield penalty (Chimenti et al., 2006).

One suggestion for the benefit of OA is that it could allow more soil water extraction due to changing the hydrostatic pressure gradient in the leaves so that plants can extract more water from dry soil, and hence, the plants can survive longer during drought (Taiz, Zeiger, Møller, and Murphy, 2015). Specifically, the lowering of osmotic potential (OP), and consequently lowering the hydrostatic potential in the plant, would increase the hydrostatic pressure gradient between the plant and soil, and can result in additional water uptake from the soil. The few studies that have considered the increased water extraction hypothesis have focused on water uptake of plants at very low soil water contents as a result of OA during soil drying.

As discussed in Chapter 2 of this dissertation, Chimenti et al. (2006) observed a low osmotic adjustment line (LOA) suffered from more severe wilting and earlier in the drought treatment than a genetically similar high osmotic adjustment line (HOA). Chimenti et al. (2006) suggested the basis for this observed difference in response to drought might be a linkage between observed differences in leaf OA and root tip OA and overall root extension. However, the studies in Chapter 2 failed to provide evidence to support the hypothesis of Chimenti et al., (2006).

In this chapter, an alternative to the root extension hypothesis was explored. This alternate perspective was focused on the possibility that the observed difference between leaf OP of the LOA and HOA line found by Chimenti et al. (2006) and in Chapter 2 could result in differences between the two lines in the rate of soil water extraction from the soil. It is

fundamental that the rate of transpiration ( $Tr$ ) is dependent on the hydrostatic pressure gradient ( $\Delta P$ ). In equation form with a conductance ( $c$ ),  $Tr = c \cdot \Delta P$ . If there is a greater  $\Delta P$ , there is the possibility that the transpiration rate will be higher, allowing for greater photosynthesis as a result of higher stomatal conductance. This relationship indicates that a constitutive difference in leaf OP among lines could result in differences in  $Tr$  even when soil water content is fairly high. This view has apparently not been pursued to investigate the consequences in tracking temporal difference in the use of water by plants as a result of genotypic differences in leaf OP at high soil water content. The focus of this chapter was to compare the pattern of soil water use by the HOA Line and the LOA Line of Chimenti et al. (2006) to gain insight about the temporal pattern of plant water use that might be associated with their differences in leaf OP. The specific objectives were to (1) determine transpirational differences between the two lines under different VPD conditions, (2) document possible differences between the two lines in the transpiration response curves to soil drying, and (3) determine possible difference between the two lines in their rate of soil water extraction. These objectives were pursued by studies conducted under stable environmental conditions of plants grown in pots in a growth chamber and in the greenhouse.

## **Materials and Methods**

### **Plant Material**

The two lines of maize used in Chimenti et al. (2006) were the focus of this research. Chimenti et al. fully described the generation of these two lines. Briefly, 20 elite inbred maize lines originating from Argentina's flint germplasm adapted to temperate environments were phenotyped for OA during their early vegetative development. Among these 20 lines, the two extreme lines with the highest OA and lowest OA were mated. The F1 seed obtained from the

cross were sown and selfed to create a F2 population. The F2 seed were sown and also selfed. Root extension of seedlings of 141 F3 families was evaluated when subjected to a -0.8 MPa osmotic solution. The 16 F3 families having the lowest seedling extension and the 16 F3 families with the highest seedling extension were each bulked forming two breeding populations. Their subsequent field tests under rain shelters found differences between the two populations in leaf OA and soil water extraction with developing soil water deficit. Although the OP of the two lines were near equivalent at high soil water content, the lines differed in OA with soil drying. One line showed low OA (LOA) and the second line showed high OA (HOA) up to 0.54 and 0.41 MPa in their two experiments.

### **Transpiration Rate vs. VPD**

Measurement of transpiration rate over a range of vapor pressure deficit (VPD) levels was done for the two maize lines using the protocol described by (Shekoofa et al., 2016). Plants were grown in pots (100 mm in diameter and 180 mm tall) constructed out of PVC pipe. A hole was drilled in a flat end cap to allow for drainage and glued to the end of the PVC pipe. Each pot was filled with a soil substrate mix (66% pea gravel and 33% Sunshine Redi-Earth Pro Growing Mix consisting of Canadian Sphagnum peat moss 50-65%, vermiculite, dolomitic lime, 0.0001% silicon dioxide). The maize plants were sown on 13 September 2013 with 3 seeds per pot. Plants were initially grown in a greenhouse at the North Carolina State University Phytotron, Raleigh, NC under 32°C/day and 26°C/night temperatures. The plants were watered daily as needed to maintain well-watered conditions. Seven days after sowing, each pot was thinned to one plant per pot.

Once plants had developed four fully expanded leaves, four plants of each line were selected for uniformity of size to be used in the measurement of transpiration vs. VPD. These measurements were done by moving plants into a walk-in controlled environment chamber at North Carolina State University's Phytotron. Chamber temperature was set to 32°C during the day and 26°C during the night. Photosynthetically active radiation in the growth chamber was an average of 533  $\mu\text{mol m}^{-2} \text{s}^{-1}$ .

The plants were given two days to acclimate to growth chamber conditions, and then the VPD response was measured over two days. Each evening before measurements the pots were watered until dripping. The soil was then covered with aluminum foil to avoid water loss via soil evaporation. On the morning of the first day, the four experimental plants of each line were each enclosed in individual 21-L clear chambers (VPD chamber). Each VPD chamber contained a 12-V computer box fan 76 mm in diameter (Northern Tool and Equipment, Burnsville, MN) to stir the air inside the chamber to help maintain plant temperature near air temperature. Temperature and relative humidity were logged every five minutes in each VPD chamber by a data logger (Lascar Electronics, Erie, PA) mounted into the side wall of the VPD chamber. VPD was calculated from the temperature and relative humidity data.

Humidity in the VPD chambers was set to target values by varying air flow rate through the VPD chambers and occasionally pre-treating air by flowing it through a column (6.35 cm wide by 117 cm tall) of silica gel (Fisher Scientific, Fair Lawn, NJ). Low air flow rate was used to achieve low VPD and high air flow plus silica gel pretreatment resulted in high VPD. Three VPD ranges were targeted on each of the two days of measurement: low VPD (0.5–1.5kPa), medium VPD (1.5–2.5kPa), and then high VPD (2.5–4.0kPa).

On both days, after the lights were turned on each morning, plants were given 30 minutes to acclimate. Following the acclimation period, all pots were weighed to obtain their initial weights. The plants were then exposed to the low VPD level (high humidity) for one hour, and then weighed again at the end of that period. This entire process was repeated again for the medium VPD level and the high VPD level. The transpiration rate of each plant was calculated for each of the three VPD levels as the difference between the initial and final weights for each VPD exposure. At the end of the second day, total leaf area of each plant was measured using a leaf area meter (LI-1300, Licor, Lincoln, NE). Leaf area was measured so that TR could be expressed on a plant leaf area basis.

All data within the HOA Line and the LOA Line were used in the regression of transpiration rate vs. VPD. Initially a two-segment linear regression was attempted for both lines using Graph-Pad Prism Version 6.0 (GraphPad Software Inc., San Diego, CA). The two linear segments were tested to determine if their slopes were significantly different ( $p < 0.05$ ). If the slopes were different, the regression analysis yielded a breakpoint between the two linear segments as well as the slope of each segment. If the slopes were not significantly different, data from all VPD treatments were fit using a linear regression model.

### **Greenhouse Dry-Down Experiment**

The greenhouse dry-down experiment (GH) was conducted to understand how the HOA Line and the LOA Line differ in their water use under soil drying conditions. This experiment was conducted in a greenhouse at the USDA-ARS Plant Science Research Facility, Raleigh, NC (46°35' N, 39°78' W). The temperature and relative humidity of the greenhouse was recorded every five minutes using data loggers (Lascar Electronics, Erie, PA). Throughout the experiment,

the ambient weather was cool and cloudy, which led to a low VPD of approximately 1.6 kPa or less for the daylight period during most days.

Plants were grown in black pots that were 25 cm in diameter and 23 cm in height, having a volume of 11 L. The pots were filled with Black Kow (Oxford, FL) soil, which was composted cow manure-based soil high in organic matter with a fertilizer analysis (N – P<sub>2</sub>O<sub>5</sub> – K<sub>2</sub>O) of 0.5-0.5-0.5. Until the dry-down began, the plants were watered daily as needed to ensure well-watered conditions.

When the plants had developed four fully expanded leaves, the dry-down treatment was initiated, which was on 20 September 2012. The evening before the experiment, all pots were watered until dripping to make sure the soil was saturated. The pots dripped over night to pot capacity. On the following morning, each pot was enclosed in a white plastic bag that was tied around the plant stem to prevent water loss via soil evaporation. The pots were then weighed for their initial weights. Pots were organized on the greenhouse bench so that the HOA Line plants and the LOA Line plants alternated. Ten plants were grown of each line – six drought-stressed (DS) and four well-watered (WW).

Throughout the dry-down, pots were weighed daily during the midafternoon to measure water loss via transpiration. The WW plants were re-watered after weighing to return the pots to 200 g below their initial weight. This water addition avoided saturating the soil but ensured the plants did not experience drought-stress. The DS plants were not watered once the dry-down treatment started.

The measurements of pot weight were used to calculate daily transpiration rate as the difference in pot weights between consecutive days. The daily transpiration rate of each DS plant was then divided by the mean transpiration rate of the WW plants within that same line on the

same day, which gave the transpiration ratio. By calculating the transpiration ratio, daily variation due to possible daily environmental variation in the chamber was minimized. To reduce variation from plant-to-plant, the daily normalized transpiration rate (NTR) was calculated for each DS plant. NTR was calculated by dividing the daily transpiration ratio of a DS plant by the average transpiration ratio of that plant during the first three days of the dry-down experiment (before the plant started experiencing drought-stress conditions). By definition, at the start of the experiment the values of NTR for each plant were centered on 1.0.

The end point of the experiment for an individual plant was defined when  $NTR \leq 0.10$ . The final pot weight at this endpoint was used in calculating fraction of transpirable soil water (FTSW) on each day for each DS plant. The formula used for calculating FTSW was: (daily weight – endpoint weight) / (initial weight – endpoint weight). All the data for each line were combined and used to regress NTR as a function of FTSW. The regression was done for a simple exponential model ( $NTR = 1 - \exp(-K \times FTSW)$ ) using Graph-Pad Prism Version 6.0 (GraphPad Software Inc., San Diego, CA).

A two-tailed T-test was conducted to find any significant differences between the HOA Line and the LOA Line in their average water use for each day. The differences in the averages of daily water use between the HOA Line and the LOA Line were calculated for each day of the dry-down treatment and plotted on a line graph. Throughout the entire dry-down, visual wilting observations were recorded to compare wilting differences between the two lines.

### **Growth Chamber Dry-Down Experiments**

An experiment (GC-LVPD) was conducted in tall pots (104 cm tall  $\times$  10 cm diameter, 8.4 L volume) in a walk-in controlled environment chamber at North Carolina State University's

Phytotron, as described in detail in Chapter 2. The plants were grown in a loamy sand mineral soil (8.8% clay, 18.2% silt, and 73% sand). Soil was filled into the pots so they would possess uniform characteristics and volume to maintain a constant bulk density across pots. This procedure involved adding three uniform-sized scoops of soil to the pot, pausing to tap the bottom of the pots on the floor three times to allow the soil to settle, and then repeating this procedure. Three seedlings were transplanted into each of the 15 pots for each line.

Approximately 4 to 8 days after transplanting, a uniform population of plants was achieved by thinning each pot to one plant by retaining plants of consistent size across all pots.

Chamber temperature was set to 26°C during the day and 18°C during the night. The chamber was set to 14 hours of daylight and 10 hours of darkness. Photosynthetically active radiation in the growth chambers was an average of 533  $\mu\text{mol m}^{-2} \text{s}^{-1}$ . Dehumidifiers were placed in the growth chamber to regulate VPD between 1.47 to 1.76 kPa (full details are given in Table 2.1). Temperature and relative humidity of the growth chamber were logged every five minutes using data loggers (Lascar Electronics, Erie, PA).

Once plants reached the V3 stage, the dry-down experiment was initiated. The night before each experiment started, pots were watered until dripping to ensure soil water content was saturated. Twelve plants of each line were designated to be subjected to the DS treatment and three plants were designated for the WW treatment. For each experiment, plants were assigned to the DS and WW treatments by examining water loss from each plant for the first three days of the experiment. For each line, one WW plant was selected with low levels of water loss, one with mid-level water loss, and one with high levels of water loss. This was to ensure all levels of transpiration rate would be present in both the DS and WW plants. During the dry-down, WW

plants were re-watered daily to maintain a pot weight of 250 g below their initial weight. The DS plants dried as transpiration allowed.

The difference in average daily water use between the HOA Line and the LOA Line was calculated each day to identify any trend in water use through the drying cycle. A two-tailed T-test was conducted to find any significant differences between the HOA Line and the LOA Line in their average water use for each day. In addition, NTR and FTSW were calculated as explained for the greenhouse experiment. A quadratic-equation model was used to regress NTR vs. FTSW. Throughout the dry-down, visual wilting observations were recorded to compare wilting differences between the two lines.

A second growth chamber experiment was done in which the results were not confounded by root extension into deeper soil as occurred in the tall pots. In this experiment (GCDD), shorter pots 25 cm in diameter and 23 cm in height, having a volume of 11 L, were used. The soil was a loamy sand (8.8% clay, 18.2% silt, and 73.0% sand). Individual plants were established in each pot. The growth chamber was set to 26°C during the day and 18°C during the night. A VPD of 1.5 to 1.7 kPa was maintained throughout the dry-down, using humidifiers and de-humidifiers to maintain the humidity levels. The dry-down treatment began once the plants reached the V3 stage, which fell on 19 December 2016. Eleven plants per line were divided for the dry-down experiment between three WW plants and eight DS plants per line. The WW plants were watered daily based on the measured water loss to maintain the pots slightly below the initial pot weight. The DS were not watered during the dry-down experiment. The pots were weighed daily and transpiration rates calculated as described previously. The last DS plant reached the endpoint of  $NTR \leq 0.10$  on 25 January 2017.

## Results

### Transpiration Response to VPD

The first objective of this research was to determine if there are differences in transpiration rate under various VPD. Surprisingly, the two lines responded quite differently to VPD (Figure 3.1). The results for the LOA Line were described by a linear regression model while the results for the HOA Line fit the two-segment linear model. The breakpoint between the high slope under low VPD and the low slope under high VPD was at 2.20 kPa. For low VPD conditions, the slope of the HOA was 16.8 whereas the LOA Line had a slope of 11.9. This means that the rate of transpiration rate was 141% greater for the HOA Line compared to the LOA Line. However, above the breakpoint of 2.2 kPa for the HOA Line the slope for the increase in transpiration rate with VPD decreased and it was only 65% of the LOA Line. Therefore, the difference in transpiration rate between the two maize lines was dependent on environmental conditions, particularly VPD.

### Transpiration Response to Soil Drying

The objective of these experiments was to characterize the transpiration response of the LOA and HOA Lines to soil drying by examining transpiration rate (NTR) as a function of soil water content (FTSW). The first experiment was done in a greenhouse with plants grown in a soil that was essentially all organic matter. The two lines differed in their response to soil drying as the NTR of the LOA Line decreased more than the HOA Line early in the drying cycle (Figure 3.2). The K values obtained from the exponential model were smaller for the LOA Line ( $K=3.94$ ) than for the HOA Line ( $K=6.15$ ), which is an indication of the delay in slowing of the transpiration rate in the HOA Line compared to the LOA Line as the soil dried (decreasing

FTSW). However, visual wilting of the maize plants was observed to occur in the LOA Line 3 days before wilting in the HOA Line.

The experiment in the tall pots in the growth chamber (GC-LVPD) was conducted under low VPD conditions so the confounding response at high VPD was avoided in this experiment. However, the tall pots meant the roots continued extension into deeper soil in the pot. Nevertheless, the two maize lines differed substantially in their pattern of NTR with soil drying (Figure 3.3). In this case, a quadratic model fit both models ( $R^2$  values of .885 and 0.868 for HOA Line and LOA Line, respectively) although the shape of the response curves was quite different. As seen in the previous experiment, NTR of the HOA Line remained near 1.0 for the initial part of the drying cycle at values substantially greater than observed for the LOA Line. The response curve for the LOA Line was nearly linear, decreasing over the entire range with a coefficient of the quadratic term having a value of only 0.12. Visual wilting was observed in the LOA plants 4 days before wilting developed in the HOA Line. However, the endpoint of  $NTR \leq 0.1$  occurred 4 days earlier in the HOA Line than the LOA Line.

The second dry-down experiment in the growth chamber was done in large pots using mineral soil. Hence, some of the possible confounding influences of the organic soil in the greenhouse experiment and of the tall pots in the first growth chamber experiment would be eliminated. Nevertheless, the results of NTR with soil dry-down were fully consistent with the previous two experiments (Figure 3.4). The exponential equation fit the response curve of both maize lines well and the K value differed between lines ( $K = 3.92$  for the LOA Line and  $K = 5.48$  for the HOA Line). Again, NTR was sustained at a higher level in the HOA Line than the LOA Line as the soil dried (decreasing FTSW). The endpoint of  $NTR \leq 0.1$  occurred 5 days earlier in the HOA Line than the LOA Line.

## **Temporal Water Extraction**

Data from the growth chamber experiment in tall pots allowed calculation of the difference in transpiration rate between the HOA and LOA Lines on each day of the dry-down. During the initial stages of the dry-down experiment (prior to day 16), the transpiration rate of the HOA Line was greater than the LOA Line (Figure 3.5). This is consistent with the results observed at low VPD in the experiment measuring transpiration rate in response to VPD. However, after the initial water was exhausted in the soil, there was a substantial drop in the transpiration of the HOA Line relative to the LOA Line (on day 11 of the dry-down). In fact, with drier soil, the transpiration of the HOA Line was less than the LOA Line. Since Figure 3.5 is based on days after initiation of the drying cycle, the point at which the transpiration rate of the HOA Line dropped relative to the LOA Line reflects that the early higher rate of transpiration by the HOA Line. The higher transpiration rate of the HOA Line caused soil water to be drawn down more rapidly than the LOA Line, i.e. the HOA Line was at a lower FTSW. That is, the lower soil water content for the HOA Line relative to the LOA Line meant that its transpiration rate had progressed to a lower level than the LOA Line. In the GC-VLPD experiment, on day 27 of the dry-down treatment, the last DS HOA Line plant reached the endpoint in the use of transpirable soil water. The experiment for the LOA Line continued longer, having the last plant of the LOA Line reach the endpoint in the use of transpirable soil water on day 30 – three days later than the HOA Line.

## **Discussion**

Comparison of two maize lines with differing leaf OA reported by Chimenti et al. (2006) showed the lines had differences in water extraction, visual wilting, and mass accumulation.

Chimenti et al. (2006) speculated that the differences in leaf OA may have paralleled differences in root OA, which would have allowed greater root extension and deeper rooting for greater water extraction. However, the studies reported in Chapter 2 failed to support their hypothesis. That is, in comparing the LOA and HOA Lines there were no significant differences in either root-tip OP nor nodal or seminal root lengths. However, Chapter 2 did confirm that the HOA Line possessed lower (more negative) leaf OP than the LOA Line. In this chapter, these two lines of maize were analyzed to compare their differences in water use, to explore alternate hypothesis to explain the difference in delayed wilting and yield advantage of the HOA Line in the experiments of Chimenti et al. (2006).

The first objective of this research was to determine transpirational differences between the two lines under different VPD conditions. Differences in water use under different VPD levels were examined. Under low VPD conditions, the HOA Line had greater transpiration rates than the LOA Line (Figure 3.1). This observation was supported in Figure 3.5 showing directly the greater transpiration rate of the HOA Line as compared to the LOA Line in the early phases of the dry-down experiment when soil water was not limiting. These observed differences in transpiration rate in the two different experiments is consistent with the lower leaf OP (more negative) for the HOA Line (results from Chapter 2) under low VPD conditions resulting in a greater hydrostatic potential gradient between the soil and leaves in the HOA Line as compared to the LOA Line.

The second objective of this research was to document possible differences between the two lines in the transpiration response curves to soil drying. The temporal dynamics of the soil water extraction differences between the HOA Line and the LOA Line were analyzed in three separate experiments. In the greenhouse experiment, plants were grown in a very high percent

organic matter soil. Rather than the conventional two-linear segment model response of NTR to soil drying, these results were well described by the exponential response. This exponential response is consistent with the results of Wahbi and Sinclair (2007) for maize grown on an artificial soil mixture.

The results of the exponential model was that the K value for the LOA Line (3.94) was much lower than the K value of the HOA Line (6.15) (Figure 3.2). That is, as the soil dried in the drought cycle, NTR of the LOA Line decreased more than the HOA Line early in the drying cycle (Figure 3.2). This difference in the transpiration response to soil drying was repeated in the other two experiments (Figures 3.3 and 3.4). These results are fully consistent with the hypothesis that the lower OP of the HOA line allowed for a greater rate of transpiration, even under high soil water content. An important consequence of the difference in OP of the two lines is that the hydrostatic pressure of the HOA Line would be greater than the LOA Line at the initial stages of the soil drying cycle. The visual consequences of higher hydrostatic pressure in the HOA Line would be delayed wilting. In fact, in the two experiments where such observations were recorded, leaf wilting was delayed in the HOA Line relative to the LOA Line by 3 days in the greenhouse experiment (GH) and 4 days in the tall pot growth chamber experiment (GC-LVPD).

The results of the experiments showed that the higher transpiration early in the drying cycle (Figure 3.5) caused the water to be used more rapidly. The HOA Line reached the endpoint in the use of transpirable soil water by day 27 while the LOA Line reached the endpoint on day 30 of the dry-down (GC-LVPD). This difference in the duration of the soil drying cycle appears to be a result of slower use of water earlier on in the dry-down by the LOA line that allowed it to have more water remaining in the soil for it to use later in the dry-down. Since the HOA Line did

not demonstrate this technique to extend the drying cycle, the HOA Line may be more vulnerable to long-term drought compared to the LOA Line.

In summary, the failure to observe differences in leaf OA between the two lines indicate that OA is not the reason for the differences between the two lines in water extraction, visual wilting, and mass accumulation. This is unexpected since the benefit of OA is that it could allow more soil water extraction due to changing the hydrostatic pressure gradient in the leaves and by extracting more water from dry soil, plants can survive longer during drought. Instead of OA, constitutive differences in leaf OP under low VPD conditions may be causing the differences between the HOA Line and the LOA Line. As the HOA Line has lower OP in its leaves, this allows the HOA Line to obtain water more readily during the early phases of water extraction – even to the point of having delayed wilting. This is due to an increase in transpiration rate in the HOA Line compared to the LOA Line. Since the HOA Line transpired more water at the start of the dry-down treatment, it used its soil water resources quicker than the LOA Line. The LOA Line had a higher leaf OP (less negative), which was associated with a lower transpiration rate, and therefore used its soil water resources slower than the HOA line throughout the early stages of the dry-down. Because of this lower transpiration rate, the LOA Line was able to have an extended dry-down cycle longer than the HOA Line under drought conditions. Sustained soil drying favors the LOA Line with its slower use of soil water.

Considering the effects of the leaf OP differences under low VPD conditions between the two lines, there still seems to be no clear-cut advantage to either line as drought develops since the outcome depends both on the environment and the duration of the drought events. The HOA Line has a disadvantage under drought conditions because it uses its soil water resources faster in the drought, leaving not enough water to survive on later in the dry-down. Although the LOA

Line has the advantage of slower water use leading to longer survival, the LOA Line has the disadvantage of earlier wilting and a lower rate of shoot mass increase. Looking back to the study of Chimenti et al. (2006), in their Experiment 1, the drought-stress seems to have been fairly moderate, with the shoot mass decreasing by only 9.5% for the HOA Line and 38.1% for the LOA Line. In their Experiment 2, which appears to have imposed a more severe drought, the shoot mass decreased by 36.5% in the HOA Line and by 44.6% in the LOA Line. The lessening of differences in Experiment 2 may have been even less, or even reversed, in an even greater severity of stress was imposed in an experiment.

### Chapter 3 Figures

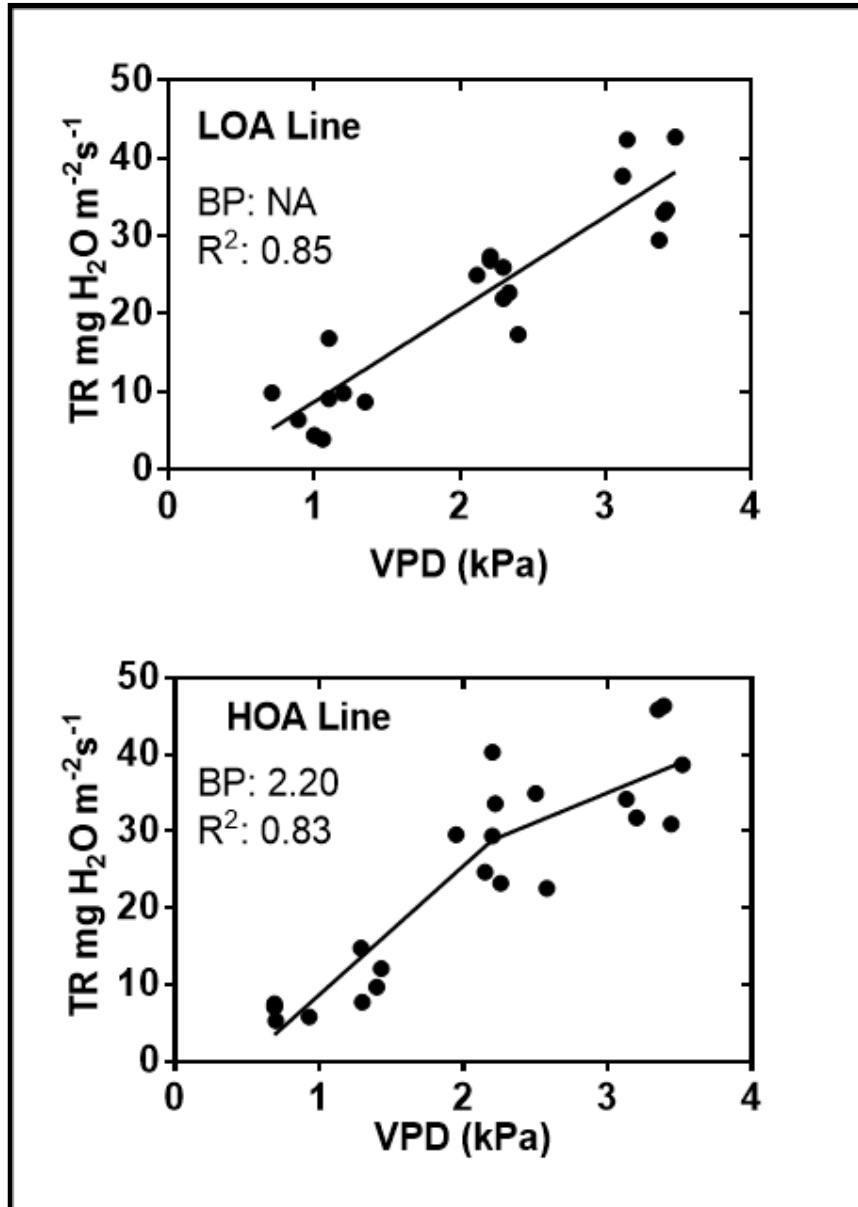


Figure 3.1: Transpiration rate ( $\text{mg H}_2\text{O m}^{-2}\text{s}^{-1}$ ) under well-watered conditions of the LOA Line (top panel) and the HOA Line (bottom panel) exposed to increasing VPD levels under controlled conditions in a growth chamber. Each data point represents one plant. For the LOA Line, a linear regression model was fit. The equation of the LOA Line is:  $Y=11.90 \cdot X - 3.20$ . For the HOA Line, represented by the two segments, the regression analysis generated a breakpoint at a VPD of 2.20 kPa between the two linear segments, which shows a linear decrease in the transpiration rate. The equation for the linear regression of the HOA Line is:  $Y=16.81 \cdot X - 8.12$ . For the second line (right side) of the HOA Line, the regression equation is:  $Y=7.72 \cdot X + 2.20$ .

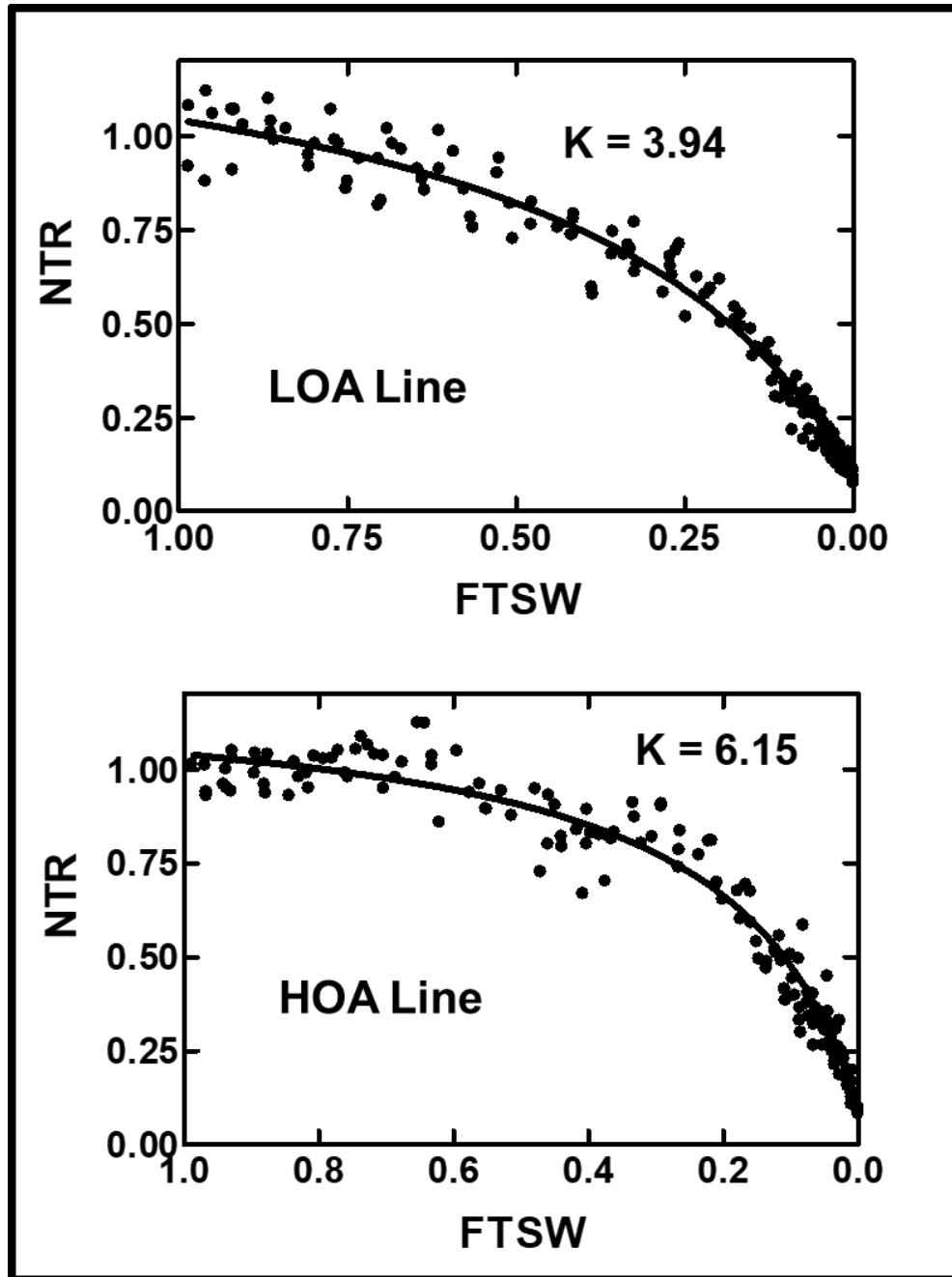


Figure 3.2: Exponential model was fit for normalized transpiration rate (NTR) versus fraction of transpirable soil water (FTSW) for the LOA Line (top panel) and the HOA Line (bottom panel). Each data point represents results for one day for each plant as the drought-stress progresses during the GH experiment.

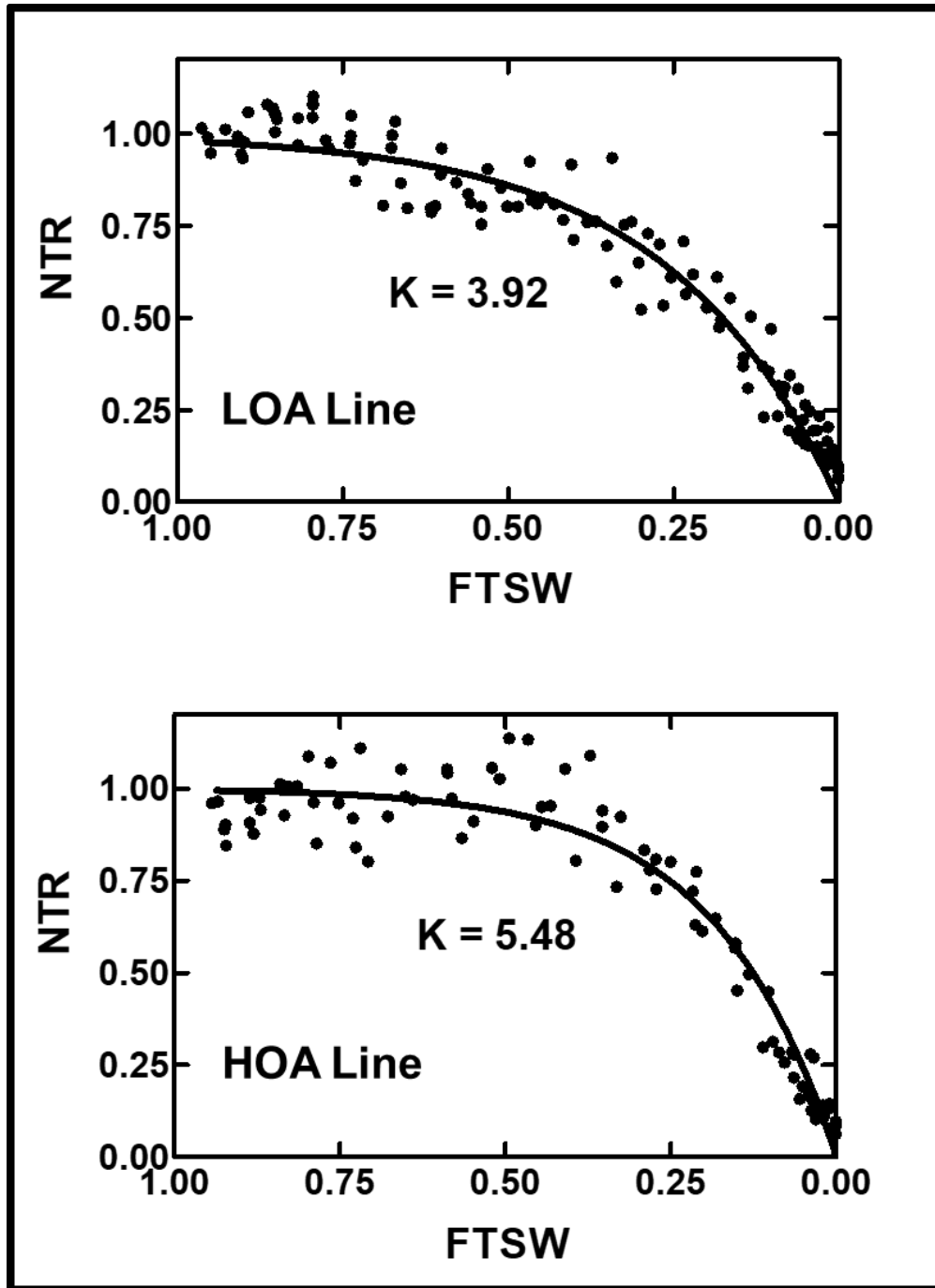


Figure 3.3: Exponential model was fit for normalized transpiration rate (NTR) versus fraction of transpirable soil water (FTSW) for the LOA Line (top panel) and the HOA Line (bottom panel). Each data point represents results for one day for each plant as the drought-stress progresses during the GH experiment.

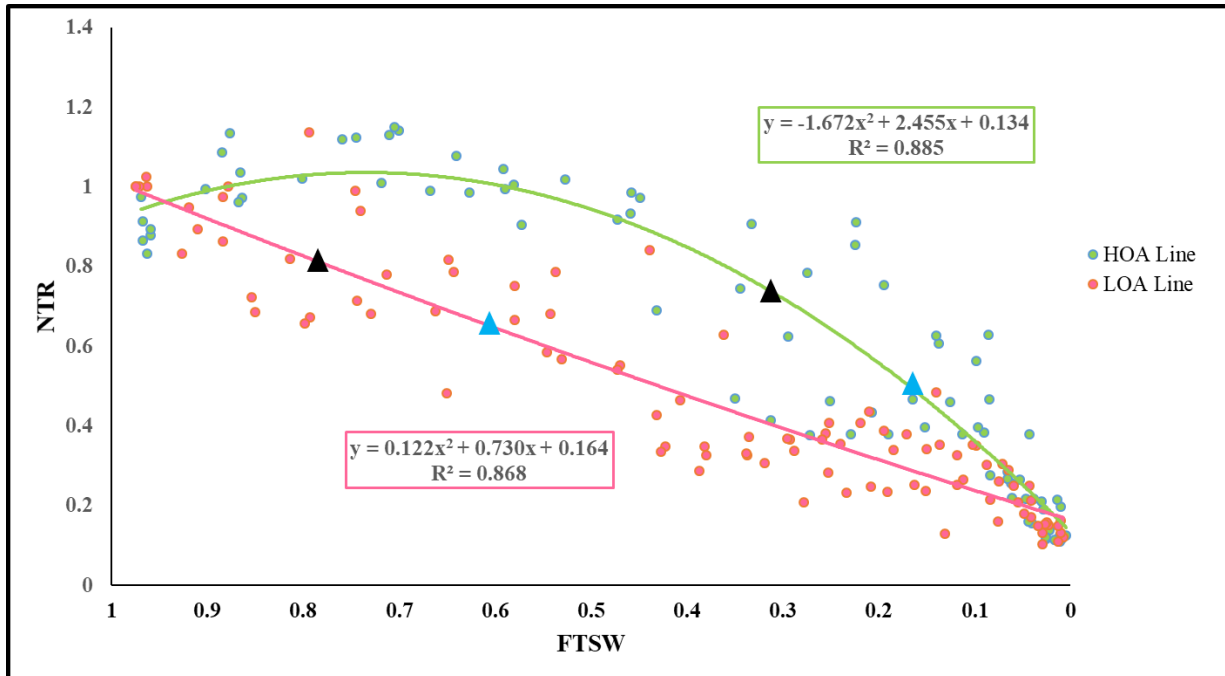


Figure 3.4: GC-LVPD: NTR vs. FTSW. Quadratic model was fit for normalized transpiration rate (NTR) versus fraction of transpirable soil water (FTSW) for the LOA Line (pink) and the HOA Line (green).the quadratic modal equations are boxed with the associated colors. Each data point represents results for one day for each plant as the drought-stress progresses during the GC-LVPD experiment. The black triangle on each line represents when that line initially began to wilt. The blue triangle on each line represents when that line began to show severe wilting phenotype.

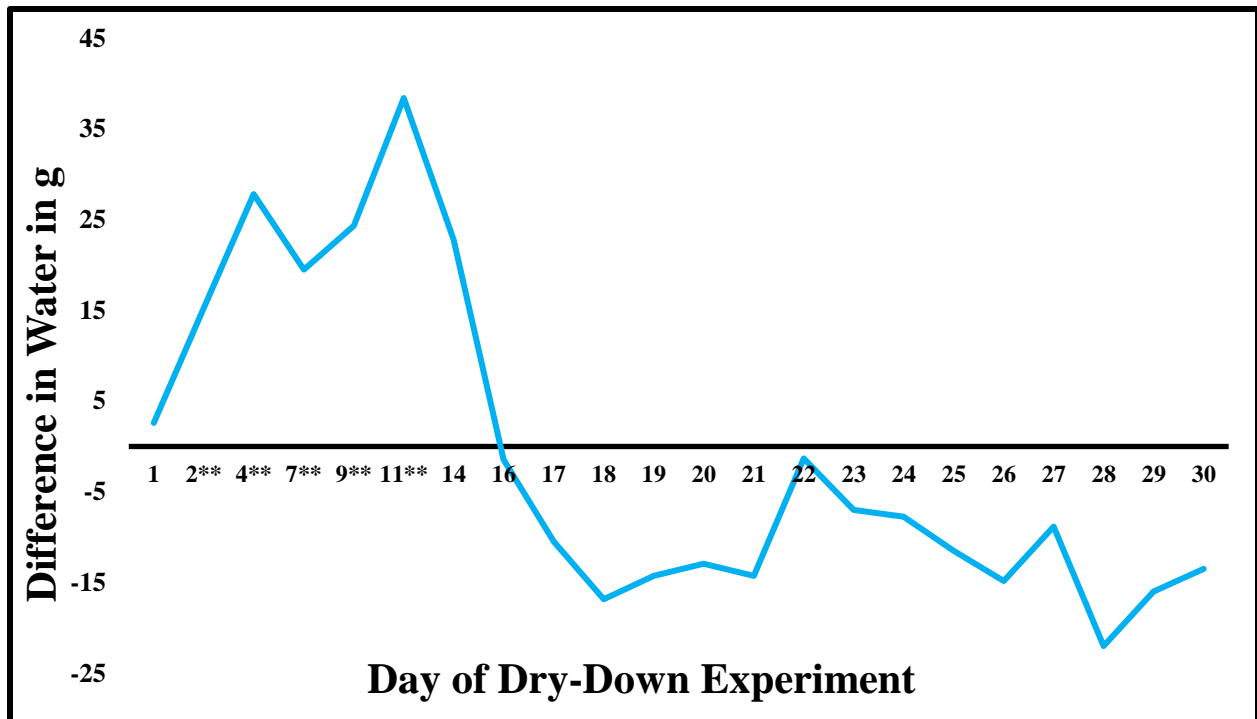


Figure 3.5: Difference in Average Water Use between the HOA Line and LOA Line. The line graph above shows differences in the averages of daily water use (in grams) between the HOA Line and the LOA Line throughout the dry-down treatment. Anytime the data is above 0, this shows that the HOA Line extracted more water that day than the LOA Line. Anytime the data is below 0, this shows that the LOA Line extracted more water the HOA Line that day. The HOA Line extracted more water than the LOA Line from the soil in the first 16 days of dry-down cycle. After day 16, the LOA Line extracted more soil water than the HOA Line. The asterisks (\*\*) along the X-axis indicate significant differences with the  $p\text{-value} \leq 0.05^{**}$ .

## **CHAPTER 4: Understanding How University Professors Teach about Climate Change**

### **Introduction**

Why do reasonable people doubt science? This is a question asked by journalists, scientists, researchers, and university professors across the U.S. in response to confusion on scientific issues outside of the scientific community, such as climate change. In October 2015, researchers from the Yale Program on Climate Change Communication released a report titled “Climate Change in the American Mind,” which shared that approximately 67% of Americans think global warming is occurring (Leiserowitz et al., 2015). That same study reported that of those 67% of Americans, approximately half of those believe climate change is caused by human activity (Leiserowitz et al., 2015). The same study showed only one in ten Americans understand that over 90% of climate scientists strongly believe that human activity is causing global warming (Leiserowitz et al., 2015). Members of the Intergovernmental Panel on Climate Change (IPCC) and other scientists agree with high certainty that global climate change is caused by human activity (Intergovernmental Panel on Climate Change, 2007; Oreskes, 2005).

Even though the science community is confident about climate change and its causes, the general U.S. public is not. As mentioned in Chapter 1 of this dissertation, public school systems do not give a high priority to teaching climate change (Sharma, 2012). Perhaps the low priority given to climate change education derives from teachers having the lack of preparedness in climate change from their university careers (Aalderen-Smeets, Molen, and Asma, 2011). In a study conducted with 1,500 public middle- and high-school teachers, less than half of the teachers reported that they had any formal climate science instruction in their university career

(Plutzer et al., 2016). Little literature is available on climate change education in the U.S. university system.

### **Statement of the Problem**

Many people state that climate change is the greatest challenge facing our world today (Battisti and Naylor, 2009). If this is the case, helping more people understand climate change is a crucial task for the university, no matter how difficult it may be to communicate and understand. In the book, *Education's End: Why our Colleges and Universities have Given up on the Meaning of Life*, Kronman (2007) states that university programs should introduce and lead students to accept the truths from modern science (Kronman, 2007).

“The Pathway to the Future” is the ten-year strategic plan of North Carolina State University (NC State), which is comprised of five goals. Goal 3 is to “enhance interdisciplinary scholarship to address the grand challenges of society” (NCSU, 2011, p. 8). One of these grand challenges in climate change. Research at NC State needs to involve not only the science of climate change, but also the educational practice of better teaching climate change science to students. This involves first understanding how university professors think about climate change, as well as understanding which climate change content is taught at the university and which teaching methods are used to teach this content.

### **Purpose of the Study**

The purpose of this research was to understand how university professors teach undergraduates about climate change. The specific research objectives were to:

1. Understand the professors' views on climate change,

2. Explore which climate change-related content they teach in their courses, and
3. Understand which teaching methods they use to teach undergraduates about climate change.

## **Theoretical Framework**

The theoretical framework used to guide this study was designed to explain teachers' attitudes toward science. This theoretical framework introduced an elaborate, comprehensive theoretical framework related to primary teachers' attitudes towards science. Throughout this dissertation this theoretical framework will be referred to as the "Attitudes Toward Science Theoretical Framework (ATSTF)." The ATSTF consists of three main components: cognitive beliefs, affect, and perceived control (Aalderen-Smeets, Molen, & Asma, 2011).

The cognitive beliefs portion of the framework refers to teachers' opinions and beliefs regarding three components:

1. "the relevance of science and science education,"
2. "beliefs about the relative difficulty of teaching science," and
3. "gender stereotypical beliefs regarding science and science teaching" (Walma, Molen, & Aalderen-smeets, 2013).

As for perceived relevance, people consider science important and perceive it relevant when they consider it "important for their personal lives, for society, for prosperity, or for health" (Aalderen-Smeets et al., 2011, p. 164). As for perceived difficulty of teaching science, this relates to the fact that people will consider how difficult the science topic is in relation to other fields of study. The third component of cognitive beliefs refers to what the person perceives to be

differences of gender roles between men and women related to their abilities to learn and teach science (Aalderen-Smeets et al., 2011).

Affect also plays an important role in the ATSTF. The affective states portion of this theoretical framework is broken into two components: enjoyment and anxiety. Enjoyment refers to the person's enjoyment of science and enjoyment of teaching science. Anxiety refers to the person's anxiety towards science and towards teaching science (Walma et al., 2013). Feelings of enjoyment in teaching science or feelings anxiety about teaching science are important to consider regarding a teacher's willingness to teach a science topic.

The third component of the ATSTF is perceived control. Perceived control refers to the amount of control teachers feel they have over the science they teach. Perceived control is comprised of two aspects in the ATSTF: self-efficacy and context dependency (Aalderen-Smeets et al., 2011). Self-efficacy refers to the level of control the teacher feels they have internally regarding their capacity to teach the scientific topic. Context dependency refers to how dependent they are to outside support (Walma et al., 2013). The presence, or absence, of contextual support is important while teachers are deciding to teach science topics (Aalderen-Smeets et al., 2012). Contextual support could include collegial support, resources, time allowed in the curriculum, and the level of effort needed to prepare to teach the topics (Appleton & Kindt, 2010). Cognitive beliefs, affective states, and perceived control lead to behavioral intention, which then leads to behavior, such as teaching climate related science topics (Aalderen-Smeets et al., 2011). See Figure 4.1 for a visual explanation of the ATSTF (Aalderen-smeets et al., 2011, p. 176).

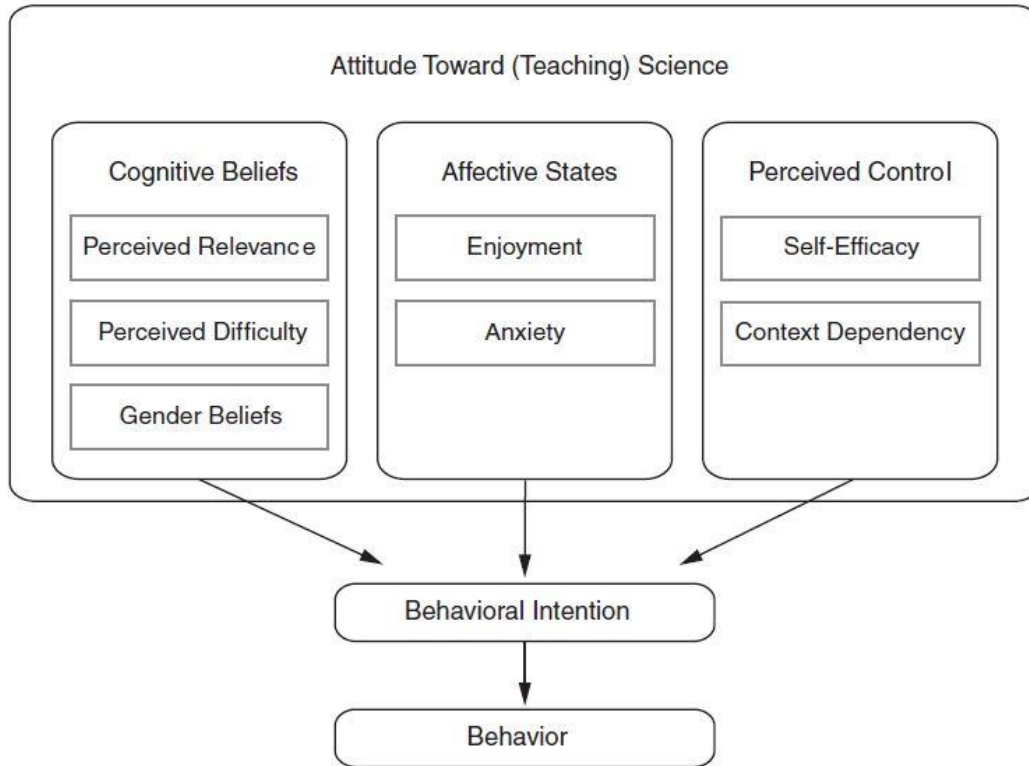


Figure 4.1: Proposed Theoretical Framework for Teachers' Attitudes toward (the Teaching) of Science (Aalderen-smeets et al., 2011, p. 176)

Although all of these components create a framework intended to look at primary teachers' attitudes towards teaching science, the ATSTF also relates to this study focusing on university professors. In relation to this study, university professors could be willing to teach about climate change based on their perceived importance of the subject, perceived difficulty of the subject, enjoyment or anxiety in teaching the topic, and their self-efficacy and support. These dimensions could also impact which content, and to what depths, they would teach related to climate change.

## **Methodology**

### **Research Design**

This study was a descriptive study utilizing survey research methodology. Quantitative research is research “which the investigator attempts to clarify phenomena through carefully designed and controlled data collection and analysis” (Fraenkel & Wallen, 2006, p. G-7). Survey research is a type of quantitative research which tries to “obtain data from members of a population (or a sample) to determine the current status of that population with respect to one or more variables” (Fraenkel & Wallen, 2006, p. G-8). This survey research possessed both descriptive and correlational characteristics and methods. Descriptive studies describe existing conditions without analyzing relationships between variables” (Fraenkel & Wallen, 2006) while correlational research studies involve “collecting data in order to determine the degree to which a relationship exists between two or more variables” (Fraenkel & Wallen, 2006, p. G-2; Gall, Borg, & Gall, 1996).

### **Population**

Purposeful sampling (Patton, 2015) was used in this study, which is when a nonrandom sample is chosen because those people selected possess the information needed (Fraenkel & Wallen, 2006). The population of this research study is comprised of NC State faculty who have taught an undergraduate course that contained one of the following phrases in their course description: “climate change,” “global warming,” and/or “environmental impact.” Twenty-six undergraduate courses met these criteria out of the 6,905 active undergraduate courses at NC State. Seventeen courses were listed under the phrase “climate change,” two under “global warming,” and seven under “environmental impact.” This population included faculty members

who taught one of these 26 courses at NC State. Faculty members were selected that have taught these courses within the last three times each of these courses were offered ( $N = 32$ ). This population was selected because they recently taught undergraduates about climate change and would therefore be able to give insight on the content and teaching methods used.

### **Survey Instrument Development**

A cross-sectional survey questionnaire was administered in this research. Cross-sectional surveys collect information at one point in time, compared to longitudinal surveys, which collect data over different points of time to study changes over time (Fraenkel & Wallen, 2006). The questionnaire was created by the researcher and then reviewed for errors and improvements by a panel of experts. The panel of experts consisted of four professors whose research programs focus on science communication and/or climate change education – two from NC State, one from the University of North Carolina at Chapel Hill, and one Auburn University.

After feedback was received from the panel of experts, improvements were made to the questionnaire. The questionnaire was then piloted with NC State University faculty members ( $n = 24$ ), all of which have teaching appointments in the Department of Plant and Microbial Biology or the Department of Horticultural Science. This group was chosen to pilot this survey because none of them were in the target population of the actual study, but they all teach courses that could possibly contain climate change content. The pilot faculty members were asked to make note of any confusing questions or errors. These were reported to the researcher and additional improvements were then made to the questionnaire.

## Reliability and Validity

As written above, a panel of experts consisting of four faculty members specializing in science communication and/or climate change education reviewed all measures of the survey instrument to provide content validity. The panel of experts reviewed all instrument questions to ensure they measured what they intended to measure.

Among all of the questionnaire items, three constructs arose by having a Cronbach's alpha greater than 0.7. A Cronbach's alpha from 0.7 to 0.8 is acceptable, 0.8 to 0.9 is good, and 0.9 and above is excellent (Gliem & Gliem, 2003). Table 4.1 shows the Cronbach's alphas for each of the three constructs.

Table 4.1: Reliability Statistics for All Three Constructs

Construct Name	Cronbach's Alpha	N of Items
Climate Change Policy Content	0.88	3
Impacts of Climate Change Content	0.93	12
Greenhouse Effect Content	0.95	10

## Data Collection

Prior to conducting any data collection, Exemption Status was sought and granted by NC State's Institutional Research Board (IRB). After receiving IRB approval, professors' emails were obtained through NC State's Registration and Records Office via a Data Request during the spring of 2018.

The survey was administered via Qualtrics, an online survey software. As recommended by Dillman, Christian, and Smyth (2014), an initial invitation email was sent to all potential

participants explaining the study. In this initial email was a link leading to the detailed consent form and then to the survey. In the invitation email, each potential participant was given a numerical code, which was mandatory for the participant to enter into their survey. This code allowed the researcher to track who responded early, late, and not at all. Also recommended by Dillman (2014), an incentive was provided to increase the response rate. Of those that completed the survey, one participant was randomly selected and given a \$25.00 gift card to a Raleigh, NC-area coffee shop of their choice. All faculty that had not yet completed the survey were sent follow-up reminder emails every five days for 25 days. All faculty that completed the survey were sent thank-you emails and one was chosen for and awarded the \$25.00 coffee shop gift card.

Thirty-two professors were asked to participate in this survey ( $N = 32$ ). Of those 32, one professor retired in 2014 and felt they had been out of the profession too long to effectively participate in the study. Two other professors replied saying that either they have not taught any of these courses or that their course does not related to climate change, so therefore they do not feel they should participate in this study. Excluding those three professors, this left 29 professors to potentially participate in this study ( $n = 29$ ) and constituted the final study frame. Of those 29 professors, 24 completed the survey, which left an 82.76% response rate. All data were downloaded from Qualtrics onto the researcher's computer and stored anonymously – the participant numerical codes or email addresses were not stored with the participants' responses.

In efforts to increase the response rate, phone calls were made to non-respondents (Dillman, Smyth, & Christian, 2008). None of the non-respondents answered the phone or returned the phone calls in response to the voicemails. After no success from the phone calls, to handle for nonresponse error, early responders were compared with late responders for variables

of interest (Miller & Smith, 1983). As Miller and Smith (1983) noted, “research has shown that late responders are often similar to nonrespondents” (p. 48). The late responders were those who responded during the last wave of responses (Lindner, Murphy, & Briers, 2001). The comparison of early and late responders used IBM SPSS Statistics 25.

### **Early Versus Late Respondents**

T-tests were used in order to compare early and late respondents to variables of interest. Only one significant difference was found between early and late respondents. The early and late respondents responded differently to the “policy construct” items, with a P-value of 0.04\*\* from the T-Test. Because of this, it is important to note that we cannot assume there will be no differences in the respondents and non-respondents related to the policy construct items.

While comparing early and late respondents, no other variables were found to be significantly different between early and late respondents, signifying that there were no other differences between early and late respondents, and therefore we can imply that there will be no other differences in the respondents and non-respondents. The early and late respondents’ comparison is shown below in Table 4.2.

Table 4.2: Comparing Early and Late Respondents with an Independent Samples T-Test

		Levene's Test for Equality of Variances		t-test for Equality of Means						
		F	Sig.	t	df	Sig. (2-tailed)	Mean Difference	Std. Error Difference	95% Confidence Interval of the Difference	
									Lower	Upper
Policy Construct	EV A <sup>a</sup>	3.72	0.07	-1.86	22	0.08	-2.18	1.17	-4.60	0.24
	EV NA <sup>b</sup>			-2.13	21.98	0.05**	-2.18	1.02	-4.30	-0.06
Impacts of Climate Change Construct	EV A	0.18	0.67	-0.90	19	0.38	-3.36	3.74	-11.19	4.47
	EV NA			-0.90	12.34	0.38	-3.36	3.71	-11.43	4.71
Greenhouse Effect Construct	EV A	0	0.99	-1.44	20	0.17	-5.33	3.71	-13.06	2.40
	EV NA			-1.35	10.21	0.21	-5.33	3.95	-14.10	3.44
Do you emphasize any of the "sides" through your teaching? (Q13)	EV A	0.17	0.69	-0.47	23	0.65	-0.31	0.78	-1.97	1.25
	EV NA			-0.48	17.92	0.64	-0.31	0.76	-1.95	1.23

Table 4.2 (continued).

In the overall design of your course, which of the following best describes your course? (Q5)	EV A	2.93	0.1	-0.04	23	0.97	-0.04	1.11	-2.34	2.26
	EV NA			-0.04	17.71	0.97	-0.04	1.09	-2.33	2.25
Which best describes the climate change content of your course? (Q15)	EV A	0.96	0.34	0.64	23	0.53	0.45	0.71	-1.02	1.92
	EV NA			0.57	11.99	0.58	0.45	0.80	-1.29	2.19
When thinking about how you teach about climate change, which of the following best describes your teaching approach? (Q18)	EV A	0.01	0.92	-0.24	22	0.81	-0.07	0.28	-0.64	0.51
	EV NA			-0.23	15.49	0.82	-0.07	0.29	-0.68	0.54

Note, <sup>a</sup>EV A stands for equal variances assumed; <sup>b</sup>EV NA stands for equal variances not assumed

## Data Analysis

Survey data were downloaded from Qualtrics and opened in Excel. In Excel, the data were copied into a new tab to be “cleaned” for use in SPSS and then all data were coded. All open-ended responses were copied to a separate tab. Data were analyzed using IBM SPSS Statistics 25. Descriptive statistics, such as frequency data, were used to determine the professors’ views on climate change, what climate change related content they teach, and what teaching methods they use to teach that climate change content.

Bivariate correlational statistics were used in this study. Spearman’s Rho rank-order correlations were calculated in IBM SPSS Statistics 25 to examine relationships between certain questions and responses from the questionnaire. Spearman Rho correlations were chosen because they measure the strength and direction of the relationship between two ordinal variables (Hauke & Kossowski, 2011).

In the correlation matrixes presented in the Results section of this chapter, correlation coefficients are displayed for each correlation. According to Hinkle, Wiersma, and Jurs (1979), a correlation coefficient between 0.00 and 0.30 has *little if any correlation* while a correlation coefficient of 0.30 to 0.50 is classified as a *low correlation*. A *moderate* correlation is when the correlation coefficient is between 0.50 and 0.70 and a *high* correlation is from 0.70 to 0.90. The strongest correlation would be from 0.90 to 1.00 and is classified as a *very high* correlation. In the correlation matrixes presented in the Results section of this chapter, \* indicates the correlation is significant at the 0.10 level and \*\* indicates the correlation is significant at the 0.05 level. Significance levels were determined via two-tailed T-Tests.

## Results

To protect confidentiality of participants, professor names were not connected with specific responses. Within the findings, the designation of P# (example: P24) is used to indicate which participant made this statement. In the example above, P24 would signify that Participant 24 made the statement in question.

Throughout the results, for questions that had a response rate of  $n = 24$ , a change of 4% represents a single respondent.

### Findings Related to Objective 1

Objective 1 of this study was to understand the professors' views on climate change. Participants were given the statement, "Climate change refers to the idea that any of the following are occurring across the world: increased temperatures, changing weather patterns, increased storm intensities, ocean acidification, melting of sea ice, or rise in sea level." They were then asked if they think climate change is happening. All of the 24 respondents ( $n = 24$ ) stated that they believe climate change is occurring. When asked how sure they were that climate change is occurring, 71% of respondents reported that they are "extremely sure" climate change is occurring, while 25% of respondents stated that they are "very sure" climate change is occurring. Only 4% of respondents stated that they are "somewhat sure" climate change is happening.

When asked what they think the cause of climate change is, 38% of respondents reported they think it is "caused mostly by human activity." Only 4% of respondents stated that they think climate change is "caused mostly by natural changes in the environment." Of all respondents, 17% of them stated that they think climate change is "caused by human activities

and natural changes in about equal parts.” Of the respondents, 4% stated “other” and specified by writing, “human activity is accelerating climate change.” Of all of the items in the questionnaire, this question asking professors about what they think the cause of climate change was the one most skipped and left unanswered; 38% of respondents chose not to respond to this question ( $n = 15$ ).

Professors were asked how important it is for a university graduate to know scientific information about climate change ( $n = 24$ ). On a scale of “1” to “5”, “1” being of little importance and “5” being very important, all professors responded with “4”s (38% of respondents) or “5”s (62% of respondents).

## **Findings Related to Objective 2**

Objective 2 of this study was to explore which climate change-related content the professors teach in their courses. In efforts to understand the climate change-related content the professors teach, professors were asked ( $n = 24$ ), “When planning the content of your course, what assumptions do you make about what your students know about climate change already before they start your course?” Respondents were able to make multiple selections from five answers. Below, Table 4.3 shows their replies with the frequency data. As Table 4.3 shows, over half of the professors assume that their students their students have heard superficial knowledge based on news reports/media. Approximately 29% of professors that responded assume that their students have no prior knowledge of climate change, while oppositely, 4% assume their students know a lot of information about climate change before day one of their course. Interestingly, no professors assume their students have knowledge of international policies that relate to climate change.

Table 4.3: Descriptive Statistics for the Professors' Assumptions of What Their Students Know About Climate Change Prior to Their Class

Item	Yes (%)	No (%)	Number of Respondents ( <i>n</i> )
I assume they have heard superficial knowledge based on news reports/media	58.3	41.7	24
I assume they understand the basic science behind climate change	41.7	58.3	24
I assume they know nothing about climate change	29.2	70.8	24
I assume they know a great amount about climate change	4.2	95.8	24
I assume they know about international policies that relate to climate change	0	100.0	24

With aim to further explore which climate change-related content the professors teach in their courses, professors were asked to report whether they show a “side” in their course related to the cause of climate change ( $n = 24$ ). Professors were specifically asked: “Sometimes people take different sides related to what causes climate change (if climate change is occurring). In your course, do you emphasize any of the “sides” through your teaching?” When answering, 38% of respondents reported that they “share that climate change is happening because of human activity.” Secondly, 33% of respondents reported that they “share that climate change is happening because of both human activity and natural changes in the environment.” Of all the respondents, 13% of them stated that they “share no side with them.” Four professors (17%) responded “other” provided comments to further explain. One wrote, “I only talk about the

importance of considering environmental impact of our buildings due to declining resources (P8).” Another professor mentioned, “Many of my students are increasingly annoyed that this topic could even be discussed: In their life the topic has been settled. They would consider this survey quite dated and they wish to move on to addressing the problem (P12).” A third professor explained, “Considering that I teach pre-modern history, climate changes in the 13th and 16th centuries are a matter of historical record, i.e., undeniable, as far as I’m concerned (P25).” No respondents said that they “share that we do not know why climate change is happening” or that they “share that climate change is happening because of natural causes in the environment. These professors quoted here teach *Structures and Materials; Energy and Environment; and From Renaissance to Revolution: the Origins of Modern Europe*.

When thinking about the content of their course, professors were asked to describe whether their climate change content was more science-related, policy-related, equally science- and policy-related, neither, or other ( $n = 24$ ). The most common response was “more climate-related science content,” having 50% of respondents choose this response. “More climate-related policy content” was selected by 17% of respondents. “Climate-related science and policy content in equal parts” was selected by 4% of respondents. Of all the respondents, 17% of them responded “neither” and 13% responded “other.” Participants that selected “other” provided the following explanations for the climate change content of their course. One participant stated that they teach “historical development of environmental destruction and pollution” (P10). Another participant mentioned that, “The focus is on materials, and GHG [greenhouse gases] and climate change are an outcome of materials selection” (P32). The other participant wrote that their climate change content is 75% science-based and 25% policy-based (P12). These professors

quoted here teach the following courses: *From Renaissance to Revolution: the Origins of Modern Europe*; *Sustainable Materials for Green Housing*; and *Energy and Environment*.

Professors were asked which climate-change related content they taught in their course, and how deep they cover those topics. This data is shown in Table 4.4 and Table 4.5.

Table 4.4: Descriptive Statistics for Impacts of Climate Change Content

Item	Depth of Coverage	Frequency (#)
Global temperature rise ( <i>n</i> = 24)	Don't teach	2
	Mention	11
	Teach	8
	Teach in Great Depth	0
Warming oceans ( <i>n</i> = 23)	Don't teach	7
	Mention	7
	Teach	8
	Teach in Great Depth	1
Shrinking ice sheets ( <i>n</i> = 23)	Don't teach	10
	Mention	8
	Teach	5
	Teach in Great Depth	0
Glacial retreat ( <i>n</i> = 23)	Don't teach	10
	Mention	8
	Teach	4
	Teach in Great Depth	1
Decreased snow cover ( <i>n</i> = 22)	Don't teach	7
	Mention	11
	Teach	4
	Teach in Great Depth	0
Sea level rise ( <i>n</i> = 23)	Don't teach	6
	Mention	8
	Teach	6
	Teach in Great Depth	3
Declining Arctic sea ice ( <i>n</i> = 22)	Don't teach	9
	Mention	8
	Teach	5
	Teach in Great Depth	0

When given the chance to provide other topics they cover in their course related to climate change, six professors responded. Professors reported that they teach the following topics in “great depth”: “better energy technologies like solar, wind, [and] geothermal to reduce fossil fuel use” (P5) and “policy responses” (P26). The following content was provided under “I teach this aspect”: “ozone hole” (P2); “loss of diversity [and] effects on the biosphere” (P5); “societal and economic crisis related to major meteorological events or changes” (P10); “health” (P11); “migration and resources” (P11); and “carbon dioxide rise” (P28).

Table 4.5: Descriptive Statistics for Greenhouse Effect Content and Policy Content

Item	Depth of Coverage	Frequency (#)
Greenhouse gases ( <i>n</i> = 24)	Don't teach	4
	Mention	5
	Teach	9
	Teach in Great Depth	6
Greenhouse Effect ( <i>n</i> = 24)	Don't teach	3
	Mention	7
	Teach	8
	Teach in Great Depth	6
How water vapor relates to climate change ( <i>n</i> = 21)	Don't teach	10
	Mention	5
	Teach	5
	Teach in Great Depth	1
How carbon dioxide relates to climate change ( <i>n</i> = 24)	Don't teach	4
	Mention	6
	Teach	11
	Teach in Great Depth	3
How methane relates to climate change ( <i>n</i> = 24)	Don't teach	8
	Mention	6
	Teach	9
	Teach in Great Depth	1
How nitrous oxide relates to climate change ( <i>n</i> = 24)	Don't teach	11
	Mention	7
	Teach	6
	Teach in Great Depth	0
How chlorofluorocarbons (CFCs) relate to climate change ( <i>n</i> = 24)	Don't teach	10
	Mention	7
	Teach	7
	Teach in Great Depth	0

Table 4.5 (continued)

How fossil fuels relate to climate change ( <i>n</i> = 24)	Don't teach	3
	Mention	10
	Teach	6
	Teach in Great Depth	5
		0
How coal relates to climate change ( <i>n</i> = 24)	Don't teach	6
	Mention	10
	Teach	3
	Teach in Great Depth	5
Solar irradiance ( <i>n</i> = 24)	Don't teach	9
	Mention	8
	Teach	5
	Teach in Great Depth	2
Intergovernmental Panel on Climate Change ( <i>n</i> = 24)	Don't teach	11
	Mention	6
	Teach	5
	Teach in Great Depth	2
Examples of international agreements on climate change ( <i>n</i> = 24)	Don't teach	11
	Mention	8
	Teach	4
	Teach in Great Depth	1
Local/state policy regarding climate change ( <i>n</i> = 23)	Don't teach	10
	Mention	7
	Teach	2
	Teach in Great Depth	4

When given the chance to provide other topics they teach in their course related to climate change, four professors responded with other content. Professors reported that they teach the following topics in “great depth”: “European and German attitudes regarding climate change” (P5); “policy history” (P11); and “outcomes of regimes” (P11). The following content was provided under “I teach this aspect”: “European and German environmental policies” (P5); “changes in agricultural methods, monocultures, [and] industrialization” (P10); and “Impacts of forest and wood materials as a carbon sink” (P32). One professor provided a content topic that they “mention,” which was “impacts of renewable energy” (P32).

Tables 4.6 and 4.7 display the Spearman Rho correlation analysis for the climate-change related content professors taught in their course. As mentioned in the Methods section of this chapter, according to Hinkle, Wiersma, and Jurs (1979), a correlation coefficient between 0.00 and 0.30 has *little if any* correlation while a correlation coefficient of 0.30 to 0.50 is classified as a *low* correlation. A *moderate* correlation is when the correlation coefficient is between 0.50 and 0.70 and a *high* correlation is from 0.70 to 0.90. The strongest correlation would be from 0.90 to 1.00 and is classified as a *very high* correlation.

Table 4.6: Correlation matrix for Impacts of Climate Change Content.

	Global temperature rise	Warming oceans	Shrinking ice sheets	Glacial retreat	Decreased snow cover	Sea level rise	Declining Arctic sea ice	Extreme events	Extreme rainfall or lack of	Extreme hurricanes	Extreme snow storms	Ocean acidification
Global temperature rise	1	<b>.80**</b>	<b>.51*</b>	<b>.55**</b>	<b>.66**</b>	<b>.70**</b>	<b>.56**</b>	<b>.66**</b>	<b>.62**</b>	<b>.44*</b>	0.29	<b>.61**</b>
Warming oceans		1	<b>.68**</b>	<b>.61**</b>	<b>.69**</b>	<b>.75**</b>	<b>.75**</b>	<b>.67**</b>	<b>.53**</b>	0.39	0.17	<b>.78**</b>
Shrinking ice sheets			1	<b>.80**</b>	<b>.75**</b>	<b>.65**</b>	<b>.87**</b>	0.38	<b>.42*</b>	0.30	0.30	<b>.67**</b>
Glacial retreat				1	<b>.88**</b>	<b>.67**</b>	<b>.66**</b>	<b>.49*</b>	<b>.60**</b>	<b>.44*</b>	0.42	<b>.60**</b>
Decreased snow cover					1	<b>.59**</b>	<b>.74**</b>	<b>.61**</b>	<b>.61**</b>	<b>.67**</b>	<b>.48*</b>	<b>.68**</b>
Sea level rise						1	<b>.49*</b>	<b>.53**</b>	<b>.65**</b>	0.29	0.21	<b>.63**</b>
Declining Arctic sea ice							1	<b>.45*</b>	0.37	0.28	0.34	<b>.67**</b>
Extreme events								1	<b>.83**</b>	<b>.56**</b>	<b>.66**</b>	<b>.46*</b>
Extreme rainfall or lack of									1	<b>.46*</b>	<b>.70**</b>	0.33
Extreme hurricanes										1	0.43	0.43
Extreme snowstorms											1	0.18
Ocean acidification												1

Note, \* indicates the correlation is significant at the 0.10 level (2-tailed). \*\* indicates the correlation is significant at the 0.05 level (2-tailed).

Table 4.7: Correlation matrix for Greenhouse Effect and Policy Content.

	GH gases	GH Effect	How water vapor relates to CC	How carbon dioxide relates to CC	How methane relates to CC	How nitrous oxide relates to CC	How CFCs relate to CC	How fossil fuels relate to CC	How coal relates to CC	Solar irradiance	IPCC	Examples of international agreements	Local/ state policy
Greenhouse gases	1	<b>.88**</b>	<b>.64**</b>	<b>.79**</b>	<b>.71**</b>	<b>.67**</b>	<b>.68**</b>	<b>.86**</b>	<b>.75**</b>	<b>.62**</b>	<b>.62**</b>	<b>.54**</b>	0.27
Greenhouse Effect		1	<b>.63**</b>	<b>.78**</b>	<b>.70**</b>	<b>.70**</b>	<b>.71**</b>	<b>.85**</b>	<b>.67**</b>	<b>.65**</b>	<b>.65**</b>	<b>.52**</b>	0.24
How water vapor relates to CC			1	<b>.71**</b>	<b>.59**</b>	<b>.86**</b>	<b>.73**</b>	<b>.47*</b>	0.34	<b>.73**</b>	<b>.42*</b>	0.19	-0.08
How carbon dioxide relates to CC				1	<b>.66**</b>	<b>.55**</b>	<b>.58**</b>	<b>.76**</b>	<b>.65**</b>	<b>.71**</b>	<b>.51*</b>	0.31	0.04
How methane relates to CC					1	<b>.42*</b>	<b>.56**</b>	<b>.69**</b>	<b>.58**</b>	<b>.55**</b>	<b>.65**</b>	<b>.48*</b>	0.15
How nitrous oxide relates to CC						1	<b>.82**</b>	<b>.41*</b>	0.31	<b>.64**</b>	0.38	0.23	-0.07
How CFCs relate to CC							1	<b>.54**</b>	<b>.50*</b>	<b>.63**</b>	<b>.42*</b>	0.34	0.06
How fossil fuels relate to CC								1	<b>.92**</b>	<b>.48*</b>	<b>.73**</b>	<b>.72**</b>	<b>.50*</b>
How coal relates to CC									1	0.31	<b>.69**</b>	<b>.77**</b>	<b>.53**</b>
Solar irradiance										1	<b>.48*</b>	0.18	-0.16
IPCC											1	<b>.87**</b>	<b>.52*</b>
Examples of international agreements												1	<b>.76**</b>
Local/ state policy													1

Note, \* indicates the correlation is significant at the 0.10 level (2-tailed). \*\* indicates the correlation is significant at the 0.05 level (2-tailed). CC stands for climate change. GH stands for greenhouse.

Tables 4.8 and 4.9 display the Spearman Rho correlation analysis between assumptions professors made about their students' knowledge of climate change prior to taking their course and the climate-change related content professors taught in their course. Table 4.10 displays the Spearman Rho correlation analysis between percentage of students per college in the professor's course and percentage of each year of undergraduate studies (grade level) in the professor's course and assumptions professors made about their students' knowledge of climate change prior to taking their course.

There were only three significant correlations found in Table 4.10. There was a high, positive significant correlation (.73) between the number of students in the course enrolled in the College of Natural Resources and the professors anticipating their students know a great amount about climate change prior to their course. There was a low, negative significant correlation (-.49) between the number of students in the course enrolled in the College of Sciences and the professors assuming their students know superficial knowledge related to climate change based on news reports and media. Finally, there was a moderate, positive significant correlation (.55) between the number of juniors in the course and the professors anticipating their students know a great amount about climate change prior to their course.

Table 4.8: Correlation matrix for Impacts of Climate Change Content and Assumptions about Student Knowledge Prior to the Professor's Course.

	Global temperature rise	Warming oceans	Shrinking ice sheets	Glacial retreat	Decreased snow cover	Sea level rise	Declining Arctic sea ice	Extreme events	Extreme rainfall or lack	Extreme hurricanes	Extreme snowstorm	Ocean acidification
I assume they know nothing about CC	<b>.48*</b>	<b>.60**</b>	0.36	0.20	0.21	<b>.56**</b>	0.22	<b>.45*</b>	<b>.41*</b>	0.30	-0.18	0.32
I assume they know a great amount about CC	0.12	-0.27	-0.22	-0.21	0.03	-0.06	-0.23	0.16	0.19	0.24	0.36	-0.21
I assume they have heard superficial knowledge based on news reports/media	0.12	-0.03	-0.07	-0.02	0.02	-0.27	-0.03	0.15	0.15	0.20	<b>.46*</b>	0.08
I assume they know the basic science behind CC	-0.32	<b>-.44*</b>	-0.35	-0.36	-0.26	<b>-.43*</b>	-0.19	<b>-.45*</b>	<b>-.56**</b>	-0.30	-0.30	-0.36

Note, \* indicates the correlation is significant at the 0.10 level (2-tailed). \*\* indicates the correlation is significant at the 0.05 level (2-tailed). CC stands for climate change.

Table 4.9: Correlation matrix for Greenhouse Effect and Policy with Assumptions about Student Knowledge Prior to the Professor's Course.

	GH gases	GH Effect	How water vapor relates to CC	How carbon dioxide relates to CC	How methane relates to CC	How nitrous oxide relates to CC	How CFCs relate to CC	How fossil fuels relate to CC	How coal relates to CC	Solar irradiance	IPCC	Examples of international agreements	Local/state policy
I assume they know nothing about CC	<b>.49*</b>	<b>.51**</b>	<b>.42*</b>	<b>.41*</b>	0.28	0.38	<b>.53**</b>	<b>.53**</b>	<b>.45*</b>	<b>.67**</b>	<b>.42*</b>	0.27	0.13
I assume they know a great amount about CC	-0.15	-0.16	-0.21	0.10	-0.26	-0.20	-0.22	-0.13	-0.07	0.00	-0.19	-0.19	-0.20
I assume they have heard superficial knowledge based on news reports/media	-0.27	-0.12	-0.33	-0.04	-0.04	-0.20	-0.02	-0.08	0.12	-0.17	0.08	0.15	0.06
I assume they know the basic science behind CC	-0.29	<b>-.46*</b>	-0.18	-0.24	-0.23	-0.31	<b>-.48*</b>	<b>-.42*</b>	<b>-.42*</b>	-0.26	<b>-.46*</b>	<b>-.41*</b>	<b>-.43*</b>

Note, \* indicates the correlation is significant at the 0.10 level (2-tailed). \*\* indicates the correlation is significant at the 0.05 level (2-tailed). CC stands for climate change.

Table 4.10: Correlation matrix for Percentage of Students per College and Percentage of Each Year of Undergraduate Studies with Assumptions about Student Knowledge Prior to the Professor’s Course.

	CALS %	CED %	CHASS %	CNR %	COE %	COS %	UC %	DN %	MGMT %	TEX %	Fresh %	Soph %	Jr %	Sr %
I assume they know nothing about CC	-0.11	-0.13	-0.14	-0.02	0.27	0.20	0.24	-0.17	-0.07	-0.15	-0.04	-0.01	-0.15	0.14
I assume they know a great amount about CC	-0.10	-0.12	-0.18	<b>.73**</b>	-0.20	-0.15	-0.25	-0.06	-0.16	-0.06	-0.16	-0.27	<b>.55**</b>	-0.05
I assume they know the basic science behind CC	0.05	0.03	-0.09	0.17	-0.31	-0.05	-0.25	-0.16	0.11	0.30	-0.13	-0.14	0.39	-0.00
I assume they have heard superficial knowledge based on news reports/media	-0.05	0.14	0.26	0.13	0.30	<b>-0.49*</b>	0.08	0.19	-0.11	0.17	0.11	-0.02	-0.20	-0.01

Note, \* indicates the correlation is significant at the 0.10 level (2-tailed). \*\* indicates the correlation is significant at the 0.05 level (2-tailed). CALS=College of Agriculture and Life Sciences; CED=College of Education; CHASS=College of Humanities and Social Sciences; CNR=College of Natural Resources; COE=College of Engineering; COS=College of Sciences; UC=University College (First-Year College); DN=College of Design; MGMT=College of Management; TEX=College of Textiles; Fresh=Freshmen; Soph=Sophomore; Jr=Junior; Sr=Senior.

### Findings Related to Objective 3

Objective 3 of this study was to understand which teaching methods professors use to teach undergraduates about climate change. First looking at their course design, professors were asked how they include climate change into their course – as a stand-alone unit (8% of respondents), an overall theme that is interwoven throughout the entire course (42% of respondents), as the entire focus of their course (4% of respondents), or other (46% of respondents). Of the 11 professors that responded “other,” here are their responses: “One section out of 9 [sections] is dedicated to this topic (P1);” “I don’t have a specific focus on climate change but I discuss a number of environmental topics that relate to it (P18);” “climate change is tangentially related to several topics covered (P19);” “climate change as tied to changes in atmospheric composition (P28);” and “interwoven into weather topics as relevant (P30).” These professors quoted here teach the following courses: *Introduction to Resource and Environmental Economics; Polymer Chemistry and Environmental Sustainability; Crop Ecology; and Introduction to Atmospheric Sciences I.*

Professors were also asked to respond to how frequently they use certain teaching methods and teaching resources to teach about climate change: never, rarely, occasionally, or frequently. Below, Table 4.6, shows their replies for each teaching method with the frequency data.

Table 4.11: Descriptive Statistics for Teaching Methods Used to Teach Climate Change

Item	How Often Method is Used	Frequency (#)
Lecture by myself (the course instructor) ( <i>n</i> = 23)	Never	1
	Rarely	1
	Occasionally	5
	Frequently	16
Lecture by a guest speaker ( <i>n</i> = 21)	Never	9
	Rarely	6
	Occasionally	3
	Frequently	3
A class “discussion” ( <i>n</i> = 24)	Never	3
	Rarely	5
	Occasionally	8
	Frequently	8
Scientific readings for outside of class-time (physical-science related) ( <i>n</i> = 23)	Never	6
	Rarely	4
	Occasionally	7
	Frequently	6
Scientific readings for outside of class-time (social-science related) ( <i>n</i> = 23)	Never	8
	Rarely	4
	Occasionally	8
	Frequently	3
Policy readings for outside of class-time ( <i>n</i> = 22)	Never	10
	Rarely	7
	Occasionally	3
	Frequently	2
Opinion readings for outside of class-time ( <i>n</i> = 23)	Never	12
	Rarely	6
	Occasionally	2
	Frequently	2

Table 4.11 (continued).

Class project related to climate change (n = 23)	Never	12
	Rarely	4
	Occasionally	4
	Frequently	3
<hr/>		
Documentaries (n = 23)	Never	11
	Rarely	6
	Occasionally	5
	Frequently	1
<hr/>		
Photos that show evidence of climate change (n = 23)	Never	8
	Rarely	4
	Occasionally	7
	Frequently	4

One professor provided other teaching methods and teaching recourses that they use in class to teach about climate change frequently: “videos, reports, graphs, and tables (P11).”

Table 4.12 displays the Spearman Rho correlation analysis for the teaching methods professors used to teach about climate change.

Table 4.12: Correlation matrix for Teaching Methods.

	Lecture by myself	Lecture by a guest	Class discussion	Physical science-related readings	Social science-related readings	Policy readings	Opinion readings	CC-related class project	Documentaries	Photos showing evidence of CC
Lecture by myself	1	<b>.44*</b>	<b>.64**</b>	<b>.65**</b>	<b>.42*</b>	0.41	0.26	0.33	0.34	0.37
Lecture by a guest		1	<b>.50*</b>	0.30	0.39	0.29	<b>.53*</b>	<b>.50*</b>	0.24	0.16
Class discussion			1	<b>.47*</b>	<b>.51*</b>	<b>.49*</b>	<b>.46*</b>	<b>.56**</b>	<b>.43*</b>	<b>.48*</b>
Physical science-related readings				1	0.31	0.21	0.26	<b>.43*</b>	0.19	<b>.45*</b>
Social science-related readings					1	<b>.66**</b>	<b>.70**</b>	0.39	<b>.46*</b>	0.40
Policy readings						1	<b>.57**</b>	<b>.53**</b>	0.40	<b>.46*</b>
Opinion readings							1	0.40	<b>.60**</b>	<b>.49*</b>
CC-related class project								1	-0.01	<b>.46*</b>
Documentaries									1	<b>.45*</b>
Photos showing evidence of CC										1

Note, \* indicates the correlation is significant at the 0.10 level (2-tailed). \*\* indicates the correlation is significant at the 0.05 level (2-tailed). CC stands for climate change.

This questionnaire also asked professors what resources they use to gather climate change information from to help them teach about climate change in their course. For each resource, they responded with how often they used that resource: never, rarely, occasionally, or frequently. Table 4.13 shows their replies for each resource with the frequency data.

Table 4.13: Descriptive Statistics for Resources Used to Help Professors Gather Information to Teach About Climate Change

Item	Yes (#)
Knowledge I already possessed ( $n = 23$ )	15
My personal research ( $n = 23$ )	13
Scientific journals ( $n = 23$ )	18
Textbooks ( $n = 23$ )	17
NASA website ( $n = 23$ )	7
National Geographic website ( $n = 23$ )	3
Intergovernmental Panel on Climate Change ( $n = 23$ )	9
EPA website ( $n = 23$ )	10
Wikipedia ( $n = 23$ )	5
Social media ( $n = 23$ )	2
Newspapers ( $n = 23$ )	7
Documentaries ( $n = 23$ )	6
Scientific TV shows ( $n = 23$ )	3

Five professors provided other resources they use to help them gather information so that they can teach about climate change in their course: “published books, articles, and reports (P11);” “guest lecturer provided materials (P12);” “primary sources from the 13<sup>th</sup> and 16<sup>th</sup> centuries (P25);” “NOAA (P28); and “...more popular science articles than actual science...(P5).” These professors quoted here teach the following courses: *Global Environmental Politics*; *Energy and Environment*; *From Renaissance to Revolution: The Origins of Modern Europe*; *Crop Ecology*; and *Green Germany: Nature and Environment in German Speaking Cultures*.

Finally, professors were asked which best describes their teaching approach when it comes to climate change: they teach only science, only emotion, science then emotion, or emotion then science. A large number of professors stated that they only teach the science aspects related to climate change (74% of respondents). Following that, 17% of respondents stated that they teach about science then emotion. The opposite, emotion then science, was used by 9% of respondents. No professors mentioned that they only use emotion to teach about climate change.

## **Discussion**

This descriptive investigation of how university professors teach about climate change yielded several important findings related to the professors’ views on climate change, what climate climate-related content they teach in their courses, and which teaching methods they use to teach undergraduates about climate change.

## Discussion Based on Objective 1

Objective 1 of this study was to understand the professors' views on climate change. As discovered from the results, professors believe climate change is occurring and they are either extremely sure or very sure about this. However, professors in less agreement about the cause of climate change.

There is not a clear consensus among professors about the cause of climate change, as some believe it is caused by human activity, some believe it is caused by natural changes in the environment, and some believe climate change is caused by human activities *and* natural changes in about equal parts. Of all of the items in the questionnaire, the question asking professors about what they think the cause of climate change is was the question most skipped and left unanswered; 38% of respondents chose not to respond to this question ( $n = 15$ ). This could indicate many possibilities; perhaps the professors were not sure about the cause of climate change, perhaps they were not confident in their opinion, or perhaps they were nervous about others knowing their opinion. If the professors' responses were anonymous instead of confidential, perhaps more responses related to the cause of climate change would have been received. Follow-up one-on-one qualitative interviews with professors could provide more in-depth information on what they think the cause of climate change is.

These university professors consider it important for university graduates to know scientific information about climate change and they think it is important for students to learn about climate change within the university. As discussed in the ATSTF, if teachers consider a scientific topic important for their own lives, society, health, or prosperity, then they consider it relevant science and hold a positive attitude about teaching it (Aalderen-Smeets, Molen, &

Asma, 2011). Perhaps professors feel climate change is relevant and important for students to learn because students learning it would lead to improvements in their own lives and society.

Perhaps this feeling of importance is a reason why these specific professors teach a course related to climate change, global warming, and/or environmental impact in the first place. A similar study should be conducted to gauge the opinions of university professors in other fields to understand if they also think it is important for university graduates to know scientific information about climate change. Also, further explorations should be conducted to see how many undergraduate students at the university are required to take one of these courses on climate change. If university professors agree that it is important for university graduates to know scientific information about climate change and they think it is important for students to learn about climate change within the university, then it climate change education should be covered in all university students' plans of work.

### **Discussion Based on Objective 2**

Objective 2 of this study was to explore which climate change-related content professors teach in their courses. While planning which climate change-related content to teach in their course, many professors assume students understand the basic science behind climate change. A smaller portion of professors assume their students have no prior knowledge of climate change before taking their course. This is important because if more professors expect their students to already know the basic science behind climate change than not, and the students do not actual understand the basic science, then this can lead to a lack of understanding related to climate change and students can feel “behind” on day one of class. If only 26 undergraduate courses at NC State officially list climate change or related terms in their course description, then students

may have not received climate change science elsewhere before taking one of these 26 courses. Future research should examine students' knowledge of climate change before taking one of these 26 courses. Then after they take the course to understand their climate change-related knowledge before the course and to understand how the course improved their knowledge of climate change.

Another assumption the majority of professors hold about their students before they take their course is that their students know superficial knowledge based on news reports and/or media. Interestingly, no professors assume their students have knowledge of international policies related to climate change. A future exploration of students' knowledge of climate change and where they gather their climate change information could help us understand if these professors' assumptions match reality.

With aim to further explore which climate change related content the professors teach in their courses, professors were asked to report whether they show a "side" in their course related to the cause of climate change. The findings indicate that not all professors share a side in class. Of those that choose to not share a side in class, perhaps they choose to do this so the students can form their own opinions from the facts. Future research is needed to explain this phenomenon. If a student takes more than one course related to climate change, global warming, and/or environmental impact, then perhaps they will hear differing sides portrayed in their courses, which could lead to confusion or it could encourage them to further study climate change on their own time.

As shown in Table 4.6, compared to other topics related to the impacts of climate change, professors do not appear to be teaching about extreme hurricanes or extreme snowstorms. Future

research should examine if professors see these events as individual short-term weather events instead of thinking of them as long-term impacts of climate change.

As revealed in Table 4.7, professors tend to teach about topics related to the Greenhouse Effect and international policy, but not local/state policy related to climate change. Why are professors not teaching about local and state policy? Perhaps professors see climate change as a larger, global problem instead of a local/state issue. This possible lack of realized-relevance is discussed in the ATSTF. If teachers consider a scientific topic important for their own lives, society, health, or prosperity, then they consider it relevant science and then they would perhaps be willing to teach it (Aalderen-Smeets et al., 2011). Perhaps professors do not see local/state policy as relevant or important for students to learn because they possibly do not think their learning it would lead to improvements in their own lives and society.

Although climate change occurs at the global and international level, choices and actions made by consumers and corporations at local and state levels contribute to climate change. As seen in Table 4.7, professors are teaching more about examples of international climate change agreements and international climate change panels – they seem to be keeping the discussion at the international level. Why are professors not “bringing home” this local and state climate change-context to their students? Does this create a mentality among students that climate change is a bigger problem that belongs to someone else instead of them and their community?

This warrants further explorations so we can understand why local/state climate change-related policy is part of the discussion that is not being taught in university classrooms. It is important that students understand how climate change is relevant to them and their community so that they do not see it as solely “someone else’s problem.” Another possibility for future research would be to examine what professors teach related to personal responsibility associated

with climate change. This study did not ask professors if they teach about personal responsibility with climate change. It would be important to learn if professors teach this aspect of climate change.

Perhaps professors are not teaching about local/state policy because they do not enjoy it or it brings them anxiety. As mentioned in the ATSTF, enjoyment and anxiety play an important role in teachers' attitudes towards teaching science (Aalderen-Smeets et al., 2011). Based on the ATSTF, another possibility for professors not covering local/state policy could be because of a lack of self-efficacy. If the professors do not feel that they have the capacity or knowledge to teach about the local/state policies, then they may avoid teaching this topic (Aalderen-Smeets et al., 2011). It could also be due to context dependency in that if teaching about local/state policies take a lot of their time and require them to dive into additional resources, they may avoid teaching this topic (Aalderen-Smeets et al., 2011).

Tables 4.8 and 4.9 show that professors' assumptions about what their students know about climate change prior to their course does not dictate what climate change-related topics they teach in their course. As seen in Table 4.10, the number of students in their course from each college does not indicate how much professors assume their students know about climate change prior to their course. It also shows that the number of students from each grade level in their course does not indicate how much professors assume their students know about climate change prior to their course. Therefore, college and grade level are not correlated to assumptions these professors make of climate change pre-knowledge of their students. It is important to note that the lack of significant correlations in Tables 4.8 and 4.9 could be due to the small sample size in this study. However, even though the sample size was small ( $n = 24$ ), all professors that officially teach undergraduate courses covering climate change were included in this research.

There were only three significant correlations in Table 4.10. There was a high, positive significant correlation (.733) between the number of students in the course enrolled in the College of Natural Resources and the professors assuming their students know a great amount about climate change prior to their course. This indicates that professors expect students from the College of Natural Resources to know about climate change before starting their course. There was a low, negative significant correlation (-.492) between the number of students in the course enrolled in the College of Sciences and the professors assuming their students have heard superficial knowledge related to climate change based on news reports and media. This indicates that professors do not assume students in the College of Sciences have heard superficial knowledge related to climate change based on news reports and media prior to taking their course. Finally, there was a moderate, positive significant correlation (.550) between the number of juniors in the course and the professors assuming their students know a great amount about climate change prior to their course. This indicates that professors expect juniors to know a great amount about climate change before they take their course. This is surprising because there was not a significant correlation related to seniors. Perhaps this is due to a low number of seniors enrolled in these courses instead of professors not expecting seniors to know a great deal about climate change.

### **Discussion Based on Objective 3**

Objective 3 of this study was to understand which teaching methods professors use to teach undergraduates about climate change. When examining how climate change is included into course design by professors, the most common is to have climate change as an overall theme interwoven throughout the entire semester. However, some professors do teach climate change as

a stand-alone unit, as the entire focus of their course, or as it relates to other topics. While teaching about climate change, the majority of professors only teach science aspects related to climate change and they do not teach about the emotional side. Of those that do teach both science- and emotion-related material, the majority of professors teach about the science first and then the emotion. A follow up study should be conducted by examining the syllabus for each of these 26 courses to better see how professors build climate change into their semester plan.

As for the specific teaching methods used to teach about climate change, the most frequently used teaching methods are: lectures by the course professor, in a class discussion, and assigning scientific readings for outside of class time that are related to physical science. The most uncommonly used teaching methods for teaching about climate change appear to be: lecture by a guest speaker, assigning scientific readings for outside of class time that are social-science related, assigning policy readings for outside of class time, assigning opinion readings for outside of class time, class projects related to climate change, and documentaries. Future research should include surveying undergraduate students about which teaching methods they feel are most effective in helping them learn about climate change. Future research should also include asking professors which lessons, activities, and assignments they feel are most impactful.

When preparing their course material and lessons about climate change, professors appear to use the following resources most commonly: knowledge they already possess, their personal research, scientific journals, and textbooks. The least commonly used resources are: the National Geographic website, Wikipedia, social media, and scientific TV shows.

## CHAPTER 5: Final Discussion, Recommendations, and Implications

### Introduction

The research presented in Chapters 2 and 3 of this dissertation involved two lines of maize (*Zea mays* L.) that have been previously studied by Chimenti, Marcantonio, and Hall (2006) – a high osmotic adjustment line (HOA) and a low osmotic adjustment line (LOA). Chimenti et al. (2006) compared the two lines of maize with diverging levels of OA to understand the value leaf OA has in increasing yield under drought-stress. In their study, Chimenti et al. (2006) saw the LOA Line wilted earlier than the HOA Line. While hypothesizing on what could be the cause of the delayed wilting in the HOA Line, Chimenti et al., (2006) mentioned that perhaps the HOA Line possessed deeper rooting as a result of OA. If the greater leaf OA of the HOA Line extended to greater OA in the root tips, then there was the possibility for greater root extension due to a greater turgor pressure. That is, the roots of the HOA Line would be able to grow deeper into the soil profile, access more soil water, and therefore have delayed and less severe wilting, compared to the LOA Line. In fact, Chimenti et al., (2006) reported that the HOA Line extracted more water than the LOA Line in both of their two field experiments. However, Chimenti et al., (2006) did not examine osmotic potential (OP) differences in the root tips of the two lines of maize nor did they measure rooting depths of the two lines.

## **Chapter 2 – Comparing Rooting Lengths and Root Osmotic Potential Between Two Lines of Maize**

The studies in Chapter 2 of this dissertation examined these two lines of maize in further detail and aimed to (1) repeat the leaf osmotic measurements as done in Chimenti et al. (2006) for additional documentation of differences in leaf OA, (2) determine whether differences in OA between the two lines occurred in the root tips during drought, and (3) determine whether there were rooting length differences between the HOA Line and the LOA Line.

In Chapter 2, it was found that under low VPD conditions, the HOA Line expressed a more negative OP compared to the LOA Line. Related to OA in the leaves, both lines failed to show significant amounts of OA occurring in their leaves. The significant differences in leaf OP between the two lines were not seen under higher VPD conditions. Related to OP in the root tips, mean root tip OP values were not significantly different between the LOA and HOA Lines. These results contradict the hypothesis of lower OP in the roots of the HOA Line. Through analyzing nodal and seminal root lengths for the two lines, no significant differences between the HOA Line root lengths and the LOA Line root lengths were shown consistently in any stage of growth.

Based on the work presented in Chapter 2, additional studies could be done to examine differences in leaf OP across various growth stages and environmental conditions. This is suggested because the leaf OP data presented in Figure 2.1 appears scattered. Also, leaf OP appeared to increase to some extent as the soil dried, which is opposite of what would occur with OA. It would be ideal to have more samples and more data points to compare. However, in order to do this, more experimental plants would need to be grown (instead of 12 DS and 3 WW per line). As these are leaf samples, taking punches from the same plants repeatedly can cause

damage and affect their photosynthetic ability and transpiration. Having more replications per line would be ideal, if the researchers have the labor and logistic capabilities to do this. It would also be ideal to have as many WW plants as DS plants. This would allow for leaf OA to be calculated by direct comparison of the two watering treatments as done in Chimenti et al. (2006) instead of how it was calculated in this dissertation. In Chimenti et al. (2006), leaf OA was calculated as the difference between the leaf OP of the WW plants and the leaf OP of the DS plants. In our experiments, OA was determined by analyzing the significance of the slope of each line of OP vs. NTR in the linear regression. Calculating OA like done in Chimenti et al. (2006) would allow us to see if the differences in our leaf OA observations and the leaf OA observations of Chimenti et al. (2006) were due to differences in how OA was calculated.

In additional experiments, nodal root tip OP should also be reexamined with more replications. Additional nodal root tip samples should be taken from DS plants all throughout the drying cycle, especially at earlier NTR values, to determine the nodal tip root OP. This should be repeated because as seen in Figure 2.2, there are not a lot of data present at higher NTR values.

Additional research studies could include examining the nodal root tip OP, seminal root tip OP, and rooting length in wide range of genetic diverse maize lines. Perhaps the hypothesis of higher root tip OP leading to longer roots which then allows for additional water uptake is relevant to other lines of maize. If that is found to be true in other lines of maize then those lines could be studied for relevance in improving drought tolerance.

### **Chapter 3 – Dynamics of Temporal Water Use in Two Lines of Maize**

Since differences in OP in the root tips, nor differences in rooting length, could explain observed differences in wilting between the two lines under drought, an alternative hypothesis

related to difference in the water use by the two lines throughout time was explored in Chapter 3 of this dissertation. Chapter 3 aimed to examine the differences in water use between the two lines as a possible alternative hypothesis for the delayed wilting in the HOA Line. The specific research objectives of Chapter 3 were to: to (1) determine transpirational differences between the two lines under different VPD conditions, (2) document possible differences between the two lines in the transpiration response curves to soil drying, and (3) determine possible difference between the two lines in their rate of soil water extraction.

In Chapter 3, it was discovered that the HOA and LOA Lines of maize respond quite differently to VPD. Under low VPD conditions and in the early part of the dry-down cycle, the HOA Line had greater transpiration rates than the LOA Line. This observed difference in transpiration rate is consistent with the lower leaf OP for the HOA Line (results from Chapter 2) under low VPD conditions having a greater hydrostatic potential gradient between the soil and leaves in the HOA Line as compared to the LOA Line. The visual consequences of higher hydrostatic pressure in the HOA Line could be delayed wilting. In fact, in the two experiments where such observations were recorded, leaf wilting was delayed in the HOA Line relative to the LOA Line by 3 days in the greenhouse experiment and 4 days in the tall pot growth chamber experiment under low VPD.

The results of the experiments in Chapter 3 eventually showed, however, that the higher transpiration early in the drying cycle caused the water to be used more rapidly and the HOA Line reached the endpoint in the use of transpirable soil water by day 27 while the LOA Line reached the endpoint on day 30 of the dry-down (GC-LVPD). Perhaps the extended time period of the drying cycle observed in the LOA Line was because a slower use of water earlier on in the dry-down allowed it to have more water remaining in the soil for it to use later in the dry-down.

Since the HOA Line did not demonstrate this water conservation technique, the HOA Line may be more vulnerable to long-term drought compared to the LOA Line. Unfortunately, the LOA Line is also not a great contender for drought tolerance. Although the LOA Line has the advantage of slower water use leading potentially to longer survival, the LOA Line has the disadvantage of earlier wilting and an increase in shoot mass decrease. Looking back to the study of Chimenti et al. (2006), in their Experiment 1, the drought-stress was more moderate, with the shoot mass decreasing by only 9.5% for the HOA Line and 38.1% for the LOA Line. In their Experiment 2, which appears to have imposed a more severe drought, the shoot mass decreased by 36.5% in the HOA Line and by 44.6% in the LOA Line.

As the findings of Chapters 2 and 3 show, these two lines of maize – the HOA Line and the LOA Line – do not appear to have outstanding drought tolerant mechanisms. Additional research with these two lines of maize should also involve drought recovery experiments. As the LOA Line survives longer than the HOA Line under drought conditions, what if a rain comes during the last part of the LOA Line's life? Studies should be done to understand how the two lines of maize recover after being re-watered after experiencing severe drought. If the LOA Line can recover well after being re-watered, and it is then capable of yielding a good quality and quantity crop, then perhaps it can still be a contender for being a valuable drought tolerant line, at least in places where drought does not typically extend past 30 days.

As of now, these two lines have been useful in expanding our understanding of how osmotic potential impacts the rate of water uptake. It is important to continue research that will help identify traits in maize and more drought tolerant lines that can improve production in dry environments. As the climate continues to change and as droughts increase and worsen, improved drought tolerant varieties are becoming ever more important.

## Chapter 4 – How University Professors Teach About Climate Change

The study in Chapter 4 aimed to understand how university professors teach undergraduates about climate change. The specific research objectives were to: (1) understand the professors' views on climate change, (2) explore which climate change-related content they teach in their courses, and (3) understand which teaching methods they use to teach undergraduates about climate change. In Chapter 4, one key finding was that professors do not appear to be teaching about local and state climate change-related policy. It is recommended for future practice that local and state climate change-related policy be built into university climate change curriculum. This is important so that students will gain a perspective on how climate change is a local issue and relates to *their* life and community.

Based on the findings of Chapter 4, a follow-up study should be conducted to better understand the professors' views of the cause of climate change. Perhaps a qualitative study with one-on-one interviews would allow for more in-depth and open conversation on this topic. Another study, similar to this one, should be conducted with university professors all across campus to see if professors of all fields also think it is important for university graduates to know scientific information about climate change.

Future research studies should also be conducted with undergraduate students. It is important to understand how many students are actually required to take one of these 26 courses on climate change. It is important to understand what percentage of undergraduate students are actually taking course work on climate change during their undergraduate career, and to what extent. For the students enrolled in one of these 26 courses, future research should examine students' knowledge of climate change before enrolling in one of the courses and then after they complete the course to understand their climate change-related knowledge before the course and

to understand how the course improved their knowledge of climate change. Future research should also be conducted with undergraduate students across campus to understand students' general understanding of climate change and where they have learned that climate change knowledge from. More specifically, future research should include surveying undergraduate students about which teaching methods they feel are most effective in helping them learn about climate change.

### **Final Thoughts**

As the researcher sought to earn a PhD in Crop Science with a Minor in Agricultural Education, this PhD dissertation followed a unique approach in that two chapters were based on plant science-research and one chapter was based on educational-research. This approach of taking on a PhD with two divergent study topics was very fruitful for the researcher. The researcher hopes to teach crop science and climate change related topics to undergraduates in the future. As the researcher hopes to teach about crop science and drought tolerance, it was crucial for her to gain hands-on experience in this first, which this PhD program allowed her to do. As the researcher plans to conduct educational research throughout her teaching career, gaining an understanding of how to complete survey research was critical for her future career, which was also accomplished through this PhD program. Overall, this PhD program has better prepared the researcher for her future career in agricultural academia.

Finally, this joint PhD program was successful largely due to the input and mentorship of two different advisors. Having one advisor as an expert in drought tolerant research and having one advisor as an expert in educational survey research was mandatory. It was important that both advisors supported each other, but in practice allowed each one to advise the chapters of

their own expertise. While each advisor gave feedback on the entire dissertation, it was helpful that each advisor did not try to each play the leader of every chapter. The drought tolerant research expert was the main advisor for Chapters 2 and 3, while the educational survey research expert was the main advisor for Chapter 4.

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## APPENDICES

## Appendix A

### IRB Exempt Status Notification

Park - 12857 - IRB Protocol assigned Exempt status Inbox x  

 IRB Administrative Office <pins\_notifications@ncsu.edu> Mar 30   

to me 

Dear Amber Beseli:

Date: March 30, 2018  
IRB Protocol 12857 has been assigned Exempt status  
Title: Understanding How University Professors Teach about Climate Change  
PI: Park, Travis Dale

The research proposal named above has received administrative review and has been approved as exempt from the policy as outlined in the Code of Federal Regulations (Exemption: 46.101. Exempt b.2). Provided that the only participation of the subjects is as described in the proposal narrative, this project is exempt from further review. This approval does not expire, but any changes must be approved by the IRB prior to implementation.

1. This committee complies with requirements found in Title 45 part 46 of The Code of Federal Regulations. For NCSU projects, the Assurance Number is: FWA00003429.
2. Any changes to the protocol and supporting documents must be submitted and approved by the IRB prior to implementation.
3. If any unanticipated problems or adverse events occur, they must be reported to the IRB office within 5 business days by completing and submitting the unanticipated problem form on the IRB website: <http://research.ncsu.edu/sparcs/compliance/irb/submission-guidance/>.
4. Any unapproved departure from your approved IRB protocol results in non-compliance. Please find information regarding non-compliance here: [http://research.ncsu.edu/sparcs-docs/irb/non-compliance\\_faq\\_sheet.pdf](http://research.ncsu.edu/sparcs-docs/irb/non-compliance_faq_sheet.pdf).

Please let us know if you have any questions.

Sincerely,

Mandy Driver  
919.515.7515  
IRB Analyst  
[ncsuirboffice@ncsu.edu](mailto:ncsuirboffice@ncsu.edu)  
NC State IRB Office

Jennie Ofstein  
919.515.8754  
IRB Coordinator  
[irb-coordinator@ncsu.edu](mailto:irb-coordinator@ncsu.edu)  
NC State IRB Office

## Appendix B

### Invitation Email

SUBJECT: Help with my PhD Dissertation Research

Hi Dr. \_\_\_\_\_,

My name is Amber Beseli and I am a graduate student working on my PhD in Crop Science with a Minor in Agricultural Education.

For my Agricultural Education research, I am looking at how university professors/instructors teach about climate change. As we think about educating our graduates about scientific issues in the world, we have an interest in specifically learning about how we educate about climate change.

You are being asked to take this survey because you teach/have taught an undergraduate course that has the words "climate change," "global warming," or "environmental impact" in the course descriptions in the NCSU course catalog.

Can you please take my survey? If you are willing, please do so **by 5:00 PM next Friday (4/13/18)**. It should take 10-15 minutes to complete, if not less.

To take the survey, follow this link to the Survey:

[https://ncsu.qualtrics.com/jfe/form/SV\\_54GtlkyYRBTZlzb](https://ncsu.qualtrics.com/jfe/form/SV_54GtlkyYRBTZlzb)

You will need a **code** to take the survey. Your numerical code is: \_\_\_\_\_

#### **"What's in it for me?" you ask...**

Personal Incentive: I know right now is the "calm before the end-of-semester storm" so if you need incentive for participating, I am giving away one \$25 gift card to the coffee shop of the winner's choice!

Professional Incentive: I am happy to provide a summary of my data to you so that you can see how others on our campus are teaching about climate change in the classroom!

We appreciate the time you will spend completing this survey as we aim to better understand how university professors teach about climate change! Thank you in advance!

Thanks so much,

Amber

## Appendix C

### Reminder Email

SUBJECT: Reminder - Help with my PhD Dissertation Research

Hi Dr. \_\_\_\_\_,

My name is Amber Beseli and I am a graduate student working on my PhD in Crop Science with a Minor in Agricultural Education.

For my Agricultural Education research, I am looking at how university professors/instructors teach about climate change. As we think about educating our graduates about scientific issues in the world, we have an interest in specifically learning about how we educate about climate change.

You are being asked to take this survey because you teach/have taught an undergraduate course that has the words "climate change," "global warming," or "environmental impact" in the course descriptions in the NCSU course catalog.

Can you please take my survey? If you are willing, this is a **friendly reminder** to please do so **by 5:00 PM next Friday (4/13/18)**. It should take 10-15 minutes to complete, if not less.

To take the survey, follow this link to the Survey:

[https://ncsu.qualtrics.com/jfe/form/SV\\_54GtlkyYRBTZlzb](https://ncsu.qualtrics.com/jfe/form/SV_54GtlkyYRBTZlzb)

You will need a **code** to take the survey. Your numerical code is: \_\_\_\_\_.

#### **"What's in it for me?" you ask...**

Personal Incentive: I know right now is the "calm before the end-of-semester storm" so if you need incentive for participating, I am giving away one \$25 gift card to the coffee shop of the winner's choice!

Professional Incentive: I am happy to provide a summary of my data to you so that you can see how others on our campus are teaching about climate change in the classroom!

We appreciate the time you will spend completing this survey as we aim to better understand how university professors teach about climate change! Thank you in advance!

Thanks so much,

Amber

## Appendix D

### Thank-You Email

SUBJECT: Thank you for helping me with my PhD Dissertation Research!

Hi Dr. \_\_\_\_\_,

Thanks so much for completing the survey related to how you teach about climate change!

We appreciate the time you spent completing this survey. This research aims to better understand how undergraduates are educated on and taught about climate change at the university so that we can provide better science education opportunities within the university.

If you have questions, feel free to contact me by email by [alwillis@ncsu.edu](mailto:alwillis@ncsu.edu).

Thanks so much,

Amber

## Appendix E

### Thank-You Email for the Gift Card Recipient

SUBJECT: Thank you for helping me with my PhD Dissertation Research! And GIFT CARD FOR YOU!

Hi Dr. \_\_\_\_\_,

You are the winner of the \$25 gift card to the coffee shop of the winner's choice! Which Raleigh coffee shop would you like your gift card for?

Thanks so much for completing the survey related to how you teach about climate change! We appreciate the time you spent completing this survey.

This research aims to better understand how undergraduates are educated on and taught about climate change at the university so that we can provide better science education opportunities within the university.

If you have questions, feel free to contact me by email by [alwillis@ncsu.edu](mailto:alwillis@ncsu.edu).

Thanks so much,

Amber

## Appendix F

### Questionnaire for Chapter 4

## How University Professors Teach about Climate Change

---

### Start of Block: Default Question Block

You are being asked to complete this survey because you have taught one or more courses that have the words "climate change," "global warming," or "environmental impact" in their course descriptions in the NCSU course catalog.

These courses include: AEC 400, ARE 336, CE 478, COM 479, CS 411, EC 336, ES 200, FLG 440, FOR 430, HI 209, MEA 213, MEA 214, MEA 321, MEA 415, MEA 425, MEA 476, PS 336, ANT 354, ARC 232, ES 300, FOR 406, HI 318, NE 404, PCC 106, PSE 220, and WPS 201.

Please respond to this survey based on the course you teach/have taught that is listed above.

Note: If you teach/have taught MORE THAN ONE of the courses above, choose the ONE of those courses that you think covers climate change the most and focus only on that ONE class throughout this entire survey.

---

### North Carolina State University INFORMED CONSENT FORM for RESEARCH

Title of Study "Understanding How University Professors Teach about Climate Change"

Principal Investigator: Amber Lynn Beseli

Faculty Sponsor: Dr. Travis Park

**What are some general things you should know about research studies?** You are being asked to take part in a research study. Your participation in this study is voluntary. You have the right to be a part of this study, to choose not to participate or to stop participating at any time without penalty. The purpose of research studies is to gain a better understanding of a certain topic or issue. You are not guaranteed any personal benefits from being in a study. Research studies also may pose risks to those that participate. In this consent form you will find specific details about the research in which you are being asked to participate. If you do not understand something in this form it is your right to ask the researcher for clarification or more information. A copy of this consent form will be provided to you. If at any time you have questions about your participation, do not hesitate to contact the researcher(s) named above.

**What is the purpose of this study?** It is important that university students are informed/taught about climate change. It is important for us to understand how university professors are teaching about

climate change at the university. By examining the professors' methods of teaching climate change, we may be able to provide other professors recommendations about how to bring scientific issues, such as climate change, into their course curriculum and be more effective at teaching it.

**What will happen if you take part in the study?** If you agree to participate in this study, you will be asked to complete an online survey via Qualtrics. You will be sent this survey link through an email in March. This survey will take approximately fifteen minutes to complete.

**Risks** There are minimal risks associated with this research.

**Benefits** There may or may not be any direct benefits for you if you participate in this research study. However, the results of this research study will help the university better understand how to teach scientific issues, such as climate change, more effectively.

**Confidentiality** The information in the study records will be kept confidential to the full extent allowed by law. Data will be stored securely in a password controlled computer. Your responses will not be stored with your names or identifying features. No reference will be made in oral or written reports which could link you to the study.

**Compensation** You will not receive anything for participating.

**What if you are a university student?** Participation in this study is not a course requirement and your participation or lack thereof, will not affect your class standing or grades at your university.

**What if you are a NCSU employee?** Participation in this study is not a requirement of your employment at NCSU, and your participation or lack thereof, will not affect your job.

**What if you have questions about this study?** If you have questions at any time about the study or the procedures, you may contact the researcher, Amber Lynn Beseli, at [alwillis@ncsu.edu](mailto:alwillis@ncsu.edu) or 704-472-6960.

**What if you have questions about your rights as a research participant?** If you feel you have not been treated according to the descriptions in this form, or your rights as a participant in research have been violated during the course of this project, you may contact Deb Paxton, Regulatory Compliance Administrator at [dapaxton@ncsu.edu](mailto:dapaxton@ncsu.edu) or by phone at 1-919-515-4514.

**Consent to Participate** "I have read and understand the above information. I have received a copy of this form. I agree to participate in this study with the understanding that I may choose not to participate or to stop participating at any time without penalty or loss of benefits to which I am

otherwise entitled.” IF YOU AGREE TO CONSENT, CLICK THE “YES” OPTION BELOW. If you do not agree to consent, please click the “No” option below and then the survey will close.

Thank you,  
Amber Beseli

- Yes, I consent to participate in this study.
- No, I do not consent to participate in this study.

*Skip To: End of Survey If North Carolina State University INFORMED CONSENT FORM for RESEARCH Title of Study “Understa... = No, I do not consent to participate in this study.*

*Skip To: If North Carolina State University INFORMED CONSENT FORM for RESEARCH Title of Study “Understa... = Yes, I consent to participate in this study.*

---

1. Please enter the numerical code provided to you in your email with this survey:

---

2. Which NCSU course(s) do you teach/have taught that cover climate change?

---

3. How important is it for a university graduate to know scientific information about climate change?  
1 being of little importance; 5 being very important

- 1
- 2
- 3
- 4
- 5

4. What percentage of your in-class time is dedicated to climate change?

[so for example, if I teach a course with 40 class sessions in a semester, and I spend 4 of those entire class sessions on climate change, I would answer 10%]

---

5. In the overall design of your course, which of the following best describes your course?

- Climate change is one stand alone unit
- Climate change is an overall theme of the course that is taught interwoven throughout the entire semester
- Climate change is the entire focus of this course
- Other \_\_\_\_\_

6. Climate change can be perceived as a political topic. How do you deal with this perceived political aspect when you teach about climate change?

---

7. When planning the content of your course, what assumptions do you make about what your students know about climate change already before they start your course?

I assume they:

- Know nothing about climate change.
- Know a great amount about climate change.
- Understand the basic science behind climate change.
- Know about international policies that relate to climate change.
- Have heard superficial knowledge based on news reports/media.

8. Which of the following content do you mention in your course related to climate change? Click how often you include each topic in your course.

	I teach this aspect in great depth (1)	I teach this aspect (2)	I mention this aspect (4)	I do not teach this (3)
Global temperature rise (a)	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Warming oceans (b)	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Shrinking ice sheets (c)	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Glacial retreat (d)	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Decreased snow cover (e)	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Sea level rise (f)	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Declining Arctic sea ice (g)	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Extreme events (h)	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Extreme rainfall or lack of rainfall (floods and droughts) (i)	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Extreme hurricanes (j)	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Extreme snowstorms (k)	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Ocean acidification (l)	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>

Other (m)

Other (n)

9. Climate change refers to the idea that any of the following are occurring across the world: increased temperatures, changing weather patterns, increased storm intensities, ocean acidification, melting of sea ice, or rise in sea level.

What do you think: Do you think that climate change is happening?

Yes

No

I do not know

*Skip To: If Climate change refers to the idea that any of the following are occurring across the world: incre... = Yes*

*Skip To: If Climate change refers to the idea that any of the following are occurring across the world: incre... = No*

*Skip To: If Climate change refers to the idea that any of the following are occurring across the world: incre... = I do not know*

10. How sure are you that climate change is happening?

Extremely sure

Very sure

Somewhat sure

Not sure at all

11. How sure are you that climate change is **NOT** happening?

- Extremely Sure
  - Very Sure
  - Somewhat Sure
  - Not Sure at All
- 

12. Assuming climate change is happening, do you think it is:

- Caused mostly by human activity
  - Caused mostly by natural changes in the environment
  - Caused by human activities and natural changes in about equal parts
  - Neither because climate change is not happening
  - If other, please specify:  
\_\_\_\_\_
-

13. Related to the question above, sometime people take different sides related to what causes climate change (if climate change is occurring). In your course, do you emphasize any of the "sides" through your teaching? If so, which one(s)?

- That climate change is happening because of human activity
- That climate change is happening because of natural changes in the environment
- That climate change is happening because of both human activity and natural changes in the environment
- That we do not know why climate change is happening
- I try not to share a "side" with them
- Other: \_\_\_\_\_

14. Which of the following content do you mention in your course related to climate change? Click how often you include each topic in your course.

	I teach this aspect in great depth (1)	I teach this aspect (2)	I mention this aspect (3)	I do not teach this (4)
Greenhouse gases (a)	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Greenhouse Effect (b)	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
How water vapor relates to climate change (c)	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
How carbon dioxide relates to climate change (d)	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
How methane relates to climate change (e)	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
How nitrous oxide relates to climate change (f)	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
How chlorofluorocarbons (CFCs) relate to climate change (g)	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
How fossil fuels relate to climate change (h)	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
How coal relates to climate change (i)	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Solar irradiance (j)	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Intergovernmental Panel on Climate Change (k)	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>

Examples of international agreements on climate change (l)

Local/state policy regarding climate change (m)

Other (n)

Other (o)

---

15. Which best describes the climate change content of your course?

More climate-related **science** content

More climate-related **policy** content

Climate-related science and policy content in equal parts

Neither

Other \_\_\_\_\_

---

16. Which of the following teaching methods/resources do you use to teach about climate change related topics in your course AND how often?

	Frequently (1)	Occasionally (2)	Rarely (3)	Never (4)
Lecture by myself (the course instructor) (a)	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Lecture by a guest speaker (b)	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
A class "discussion" (c)	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Scientific readings for outside of class-time (physical-science related) (d)	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Scientific readings for outside of class-time (social-science related) (e)	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Policy readings for outside of class-time (f)	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Opinion readings for outside of class-time (g)	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Class project related to climate change (h)	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Documentaries (i)	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Photos that show evidence of climate change (j)	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>
Other: (k)	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>	<input type="radio"/>

---

17. In your opinion, what is the best teaching method to use while teaching climate change, and why?

---

18. When thinking about how you teach about climate change, which of the following best describes your teaching approach?

- Emotion then science *[example, showing a photo of a polar bear standing on melting ice looking skinny to get an emotional reaction from the students and THEN teaching them about how warming temperatures are causing shrinking ice sheets]*
  - Science then emotion *[example, teaching about how warming temperatures are causing shrinking ice sheets and THEN showing a photo of a polar bear standing on melting ice looking skinny to get an emotional reaction from the students]*
  - Only science
  - Only emotion
-

19. What resources do you use to gather climate change information from to help you teach this topic in your course? You can choose more than one response.

- Knowledge I already possessed
- My personal research
- Scientific journals
- Textbooks
- NASA website
- National Geographic website
- Intergovernmental Panel on Climate Change
- EPA website
- Wikipedia
- Social Media
- Newspapers
- Documentaries
- Scientific TV shows
- Other(s) \_\_\_\_\_

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20. What is your undergraduate degree(s) in?

\_\_\_\_\_

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21. If you hold a Master's degree(s), what is it in?

\_\_\_\_\_

22. If you hold a Doctorate degree(s), what is it in?

---

23. What is your research area(s)?

---

24. Describe your background in climate change/climate science.

---

25. Would you be willing to answer follow-up questions about how you teach about climate change either in-person or over the phone?

If so, please list your email address below.

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Thank you for participating in this survey!

26. To show my appreciation, I am giving away one \$25 gift card to the coffee shop of the winner's choice! If you would like to be considered for this gift card, please list your email address below.

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End of Block: Default Question Block

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### Construct Key

**Lime Green** = Policy Construct

**Light Blue** = Impacts of Climate Change Construct

**Yellow** = Greenhouse Effect Construct

## Appendix G

This table below displays the course numbers, course names, number of credit hours, and the keywords under which each of the 26 courses appeared in the NC State course catalog.

Course Number	Course Name	Credit Hours	Keywords in Course Catalog
AEC 400	Applied Ecology	3	Climate Change
ANT 354	Peoples and Cultures of the Pacific	3	Global Warming
ARC 232	Structures and Materials	3	Environmental Impact
ARE 336	Introduction to Resource and Environmental Economics	3	Climate Change
CE 478	Climate Change Communication	3	Climate Change
COM 479	Climate Change Communication	3	Climate Change
CS 411	Crop Ecology	3	Climate Change
EC 336	Introduction to Resource and Environmental Economics	3	Climate Change
ES 200	Climate Change and Sustainability	3	Climate Change
ES 300	Energy and Environment	3	Environmental Impact
FLG 440 <sup>a</sup>	Green Germany: Nature and Environment in German Speaking Cultures	3	Climate Change
FOR 406	Forestry Inventory, Analysis and Planning	4	Environmental Impact
FOR 430	Forest Health and Protection	3	Climate Change
HI 209	From Renaissance to Revolution: The Origins of Modern Europe	3	Climate Change
HI 318	Environmental History of Cuba: Prehistory to the Present	3	Environmental Impact
MEA 213	Introduction to Atmospheric Sciences I	2	Climate Change
MEA 214	Introduction to Atmospheric Sciences II	2	Climate Change

MEA 321	Fundamentals of Air Quality and Climate Change	3	Climate Change
MEA 415	Climate Dynamics	3	Climate Change & Global Warming
MEA 425	Introduction to Atmospheric Chemistry	3	Climate Change
MEA 476	Worldwide River and Delta Systems: Their Evolution and Human Impacts	3	Climate Change
NE 404	Radiation Safety and Shielding	3	Environmental Impact
PCC 106	Polymer Chemistry and Environmental Sustainability	3	Environmental Impact
PS 336	Global Environmental Politics	3	Climate Change
PSE 220	From Papyrus to Plasma Screens: Paper and Society	2	Environmental Impact
WPS 201	Sustainable Materials for Green Housing		

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Note, <sup>a</sup>This course (FLG 440) is taught in German, not English.