

EVALUATING THE IMPACT OF WEED COMPETITION AND WATER STRESS ON CORN  
HYBRIDS WITH DIFFERING DROUGHT TOLERANCE

By

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## ABSTRACT

### EVALUATING THE IMPACT OF WEED COMPETITION AND WATER STRESS ON CORN HYBRIDS WITH DIFFERING DROUGHT TOLERANCE

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Water stress and weed competition are critical stressors during corn (*Zea mays* L.) development. Genetic improvements in corn have resulted in hybrids with greater abiotic stress tolerance. With the expected change in precipitation throughout the Great Lakes Region, field and greenhouse studies were conducted to: 1) evaluate weed competition and water stress impacts on drought tolerant corn hybrid performance and 2) assess water stress and weed competition impacts on weed community composition and corn hybrid performance. In the field study, weed densities were lower under reduced precipitation and the communities were more diverse. As weed density increased, there was no difference in the rate of yield loss between drought sensitive (DS) hybrid under ambient or reduced precipitation. In contrast, as weed density increased the rate of yield loss was 1.8 times greater for drought tolerant (DT) hybrid grown under ambient precipitation compared to DT hybrid grown under reduced precipitation. Furthermore, as weed density increased the rate of yield loss was 1.3 times greater for DS hybrid grown under ambient precipitation compared to DT hybrid grown under reduced precipitation. There was no difference in the rate of yield loss as weed competition increased between DT and DS hybrid grown under ambient precipitation and DT and DS hybrid grown under reduced precipitation. Results from the greenhouse study confirm field study results. Results demonstrate that reduced precipitation and increasing weed pressure decreases corn yield and impacts weed species diversity and evenness. Ultimately, integrated weed management will need to adapt to these changes for continued success under future climate scenarios.

Dedicated to:  
my parents, Bridget and Jason;  
my fiancé, Dalton;  
and my grandfathers in heaven

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## CHAPTER I

### LITERATURE REVIEW

#### Introduction

Corn (*Zea mays* L.) is an annual cereal crop that is grown primarily for animal feed (Perry, 1988), various fields of the food industry (Senti and Schaefer, 1972), and ethanol (Chien et al., 1990). Grain corn has been cultivated for thousands of years (Gwartz and Garcia-Casal, 2014) with 30% of the United States cropland planted to corn annually (USDA-NASS, 2020). In Michigan, grain corn ranked second in total harvested acres (1,610,000 acres), but first in value of production (\$959,560,000) (USDA-NASS, 2019).

Corn growth and development is categorized into two main stages: vegetative and reproductive. Vegetative growth stages encompass growth from emergence (VE) through tassel (VT) (Lee, 2007). Reproductive growth stages include six stages: silking (R1), blister (R2), milk (R3), dough (R4), dent (R5), and maturity or black layer (R6) (Lee, 2007). Growth stage identification dictates corn management including fertilizer inputs and pest control timing. Early corn hybrids were often susceptible to insects, herbicides, and diseases (Babcock et al., 1999). Current hybrids are now tolerant to many of those stressors (Babcock et al., 1999). Specifically, insect tolerant hybrids revolutionized corn production when *Bacillus thuringiensis* (Bt) toxins were first expressed in corn in 1996 by reducing insecticide applications to control lepidopteran insects (Babcock et al., 1999). Additionally, herbicide tolerance to glyphosate and glufosinate introduced in 1998 and 1997 allowed weed control to shift away from high input mechanical control tactics to predominantly low input chemical control (Babcock et al., 1999). Although improvements have been made throughout Michigan, corn growers still face many challenges. One major challenge is future climate change and implications on pest control.

## Corn - Weed Competition

Since the beginning of agriculture, farmers have had to control weeds to protect crop yields. A weed is defined as an undesired plant, growing out of place, that competes for water, light, and nutrients with the desired plant (Boehm et al., 2011). Weeds are competitive in agriculture fields because they grow fast, and reproduce quickly and easily by sexual or vegetative reproduction (Sutherland, 2004).

When corn and weeds grow within the same environment competition will occur for finite resources (Kropff and Van Laar, 1993). Weeds are most competitive when they emerge before or with the crop (Swanton et al., 2015). Specifically, the most critical time to control weeds is before the 5<sup>th</sup> leaf stage (V5) (Page et al., 2012). Not all weed species have the same impact on crop competition. Competitiveness can differ based on plant height, growth rate, life and reproductive cycles, root systems, leaf area, and seed production (Swanton et al., 2015). For example, velvetleaf (*Abutilon theophrasti* Medik) germinates throughout the summer, has large leaves, and can grow taller than corn, ultimately intercepting large amounts of light (Mitich, 1991; Scholes et al., 1995). Given these characteristics, velvetleaf densities need to be kept below three plants m<sup>-2</sup> to mitigate corn yield losses (Mitich, 1991). In contrast, green foxtail (*Setaria viridis* (L.) P. Beauv.), germinates mid to late May, has less than 1% seed viability after six years, and can easily be controlled by crop rotations (Gulden and Shirliffe, 2009; Heurd and Moncada, 2010). However, if green foxtail populations do emerge, minimal yield loss will occur if populations are kept below 13 plants m<sup>-2</sup> (Weaver, 2001).

One particularly troublesome weed in Michigan is common lambsquarters (*Chenopodium album* L.). Common lambsquarters is a C<sub>3</sub> broadleaf summer annual weed that is common in many agricultural fields (Korres et al., 2016). Without crop competition, common lambsquarters

is a prolific seed producer and can produce 75,600-150,400 seeds plant<sup>-1</sup>. While in competition with corn, common lambsquarters can produce 110-3,600 seeds plant<sup>-1</sup> (Colquhoun et al., 2001).

Common lambsquarters is competitive with many crops. In a list of the world's worst weeds by Holm et al. (1977), common lambsquarters was in the top 10 worst weeds based on approximate order of crop yield losses in the world. Previous research in sugar beet (*Beta vulgaris* L.) found that eight plants in 10 m row of sugar beets could reduce yield by 48% (Schweizer, 1983). In corn, common lambsquarters can decrease yield by 12% with 49 plants per 10 m of row (Beckett et al., 1988). Harrison (1990) reported that in soybean (*Glycine max* L. Merr.), there was a yield loss of 26 kg per kg of common lambsquarters biomass.

In addition to large seed production, common lambsquarters has a relative growth rate (RGR) ranging from 0.23-0.26 g<sup>-1</sup> day<sup>-1</sup> in varying day/night temperature regimes (17/14, 25/18, and 34/28 °C) (Percy et al., 1981). Furthermore, common lambsquarters can germinate and establish at relatively low temperatures of 13°C (Chu et al., 1978). Given these biologic features of prolific seed production, RGRs that are not dependent on temperature, and establishment at low temperatures, common lambsquarters has many competitive advantages against other weed species and crops (Fischer et al., 2004).

Chu et al. (1978) reported that in comparison to redroot pigweed (*Amaranthus retroflexus* L.), common lambsquarters germinated 6.5 times more at low day/night temperatures of 13/7°C. Early germination would allow common lambsquarters to establish before redroot pigweed, and thus take over the population. Furthermore, Chu et al. (1978) speculated that if land preparation and planting were to occur in late April or early May, in conjunction with cool wet weather, common lambsquarters could dominate other C<sub>4</sub> weed species like redroot pigweed or crops like corn.

## Herbicide Resistance

The predominant method crop producers use to control weeds is through chemical applications of herbicides (Mulwa and Mwanza, 2006). Consequently, overreliance on herbicides for weed control has increased the selection pressure for herbicide resistant weeds (Mulwa and Mwanza, 2006). Resistance in general occurs when herbicides within the same site of action (SOA) are applied over multiple years (Jasieniuk et al., 1996). Currently, 262 species have evolved resistance to 23 out of the 26 known herbicide SOAs (Heap, 2020).

In the United States, the first herbicide resistant weed, smooth pigweed (*Amaranthus hybridus* L.), was reported in Maryland in 1972 and was resistant to photosystem II inhibitors (Heap, 2020). Photosystem II inhibiting herbicides inhibit photosystem II dependent reactions during photosynthesis, the most common herbicide active ingredients in corn production are atrazine and simazine (Pfister and Arntzen, 1979). Specifically in Michigan, the first weed reported to be resistant to photosystem II inhibitors was common lambsquarters in 1975 (Heap, 2020). Another important SOA that weeds in corn production have evolved resistance to are acetolactate synthesis (ALS) inhibitors. ALS inhibitors are broad spectrum chemicals that kill both grasses and broadleaves and work by inhibiting the first enzyme in the branched amino acid pathway, acetolactate synthase (Shaner, 1999). In the United States, some of the first and most troublesome weeds reported to be resistant to ALS inhibitors were tall waterhemp (*Amaranthus tuberculatus* (Moq.) Sauer) in 1993, Palmer amaranth (*Amaranthus palmeri* S. Watson) in 1993, and smooth pigweed (*Amaranthus hybridus* L.) in 1994 (Heap, 2020). Previous research has found that tall waterhemp can reduce corn yield by 11-74% (Steckel and Sprague, 2004). Massinga et al. (2001) reported that palmer amaranth can reduce corn yield from 11-91% and smooth pigweed can reduce corn yield by 39% (Moolani et al., 1964).

Another important herbicide that revolutionized corn production was glyphosate. Prior to 1996, glyphosate usage was minor, given its lack of selectivity (Benbrook, 2016). However, in 1996, genetically engineered (GE) glyphosate resistant corn, soybean, and cotton (*Gossypium hirsutum* L.) were approved for commercial use, making it possible for glyphosate to be sprayed as a cost effective postemergent weed control method (Benbrook, 2016). According to the International Herbicide-Resistant Weed Database there are 50 weed species that have evolved resistance to glyphosate (Heap, 2020). Growers adopted glyphosate as their primary weed control method, because weeds had evolved resistance to ALS, acetyl coA carboxylase (ACCase), and triazine herbicides and glyphosate was cost effective (Heap and Duke, 2018). With the number of herbicide resistant weeds continuing to rise and subsequently weed control becoming more difficult, growers will need to implement integrated weed management strategies.

Integrated weed management is a multidisciplinary approach to weed control, utilizing the application of numerous control methods including chemical, mechanical, and cultural (Swanton and Weise, 1991). Mechanical control via pre-plant tillage eliminates early germinating weeds before the crop is planted (Boehm et al., 2011). In season cultivation practices can also help eliminate weeds until crop canopy (Boehm et al., 2011). Another option for weed control is the implementation of cultural practices. Cultural practices include row spacing, planting population density, implementing cover crops, and diverse crop rotations (Reddy and Jha, 2016). By implementing cultural and mechanical control along with chemical control, the impacts of crop-weed competition can be reduced.

## Climate Change

Current global climate change projections are expecting alterations to elements that are crucial for crop production. Global projections include elevated temperatures, increase in carbon dioxide (CO<sub>2</sub>) concentrations, and an increase in frequency of erratic precipitation events (Hatfield et al., 2011).

### Temperature

Current projections for temperature suggest that the mean global temperature will continue to increase over the 21<sup>st</sup> century (Collins et al., 2013). Collins et al. (2013) expect that an increase in the frequency, duration, and magnitude of hot extremes will occur. Among a majority of climatic modelers, it is agreed that globally mean annual surface temperature will increase by 1.5 to 4.5°C (MacCracken, 2008; Singh et al., 2011).

Since 1951 the Great Lakes Region has seen an increase in annual average air temperature of -16.5°C (GLISAP, 2017). Increasing temperature can greatly impact plant growth and phenology (Hatfield and Prueger, 2015). In general, many plant species can tolerate higher temperatures during the vegetative growth stages, but not during the reproductive growth stages (Hatfield and Prueger, 2015). However, each species responds to temperature differently throughout their lifecycle. Previous research has shown that when corn tassels were exposed to temperatures above 32°C during early reproductive growth stage (VT-R1) pollen viability is reduced (Herrero and Johnson, 1980). Further, extreme temperatures can impact rate of maturity and senescence. Hatfield and Prueger (2015) reported when corn plants were grown under 34°C for 120 days, total vegetative biomass increased by 20% compared to total vegetative biomass of plants grown under 30°C; however, grain yield decreased by 88%. From this study, Hatfield and Prueger (2015) confirmed that during the vegetative growth stages of corn, warmer daytime

temperatures do not negatively impact total vegetative biomass, but do have strong negative consequences on grain yield.

In addition, it is expected that as temperature increases, planting dates of crops might shift towards early spring; however, spring precipitation can impact this decision as well (Kucharik, 2008). A survey evaluating planting date and corn yields in the Midwest found that there was a significant ( $P < 0.05$ ) relationship between planting date and corn yield, suggesting that earlier planted corn can contribute to a yield increase of 0.06-0.14 Mg ha<sup>-1</sup> for each additional day of planting earlier from the state average (Kucharik, 2008). In conjunction with planting date changes, management practices such as crop genotype, plant density, fertilizer, tillage, and weed control, will likely shift (Long et al., 2017). Specifically, weed control may become more difficult in earlier planted corn due to a greater likelihood of re-infestation, even after a postemergent herbicide application is made, influencing the critical period of weed control (CPWC) (Williams, 2006). For example, Gower et al. (2002) reported that without herbicide application giant foxtail (*Setaria faberia* Herrm.) density was 3,420 and 2,560 plants m<sup>-2</sup> in the early and late corn plantings during a wet spring and 730 and 300 plants m<sup>-2</sup> in the early and late corn plantings during a dry year. Weed communities are also expected to change as planting dates become earlier. Weed seed germination is correlated with temperature and as temperature increases, so does planting date, shifting to an earlier planting (Schwartz-Lazaro and Copes, 2019). For example, Amaranthus species were examined under varying temperature regimes and 41% germination was reached by all species at 20°C within 14 days, as opposed to 4% germination at 5°C (Steckel et al., 2004). This also indicates that temperature fluctuation may impact weed seed dormancy, which influences germination rate (Forcella et al., 1997).

With projected climate change, weed species are expected to shift geographical locations, phenology, as well as impact invasiveness and extinction (Singh et al., 2011). Under increased temperature, weed species may experience shifts in geographical locations, resulting in the invasion of species into new locations leading to localized extinctions (Amare, 2016). Under projected climate change scenarios, growing degree days will accumulate faster resulting in weed species reaching maturity earlier in the growing season (Singh et al., 2011). Singh et al. (2011) suggests that an increase in temperatures will shift floral composition of species in higher latitudes, which may result in a change of weed species flowering, fruiting, and seed dormancy.

### Carbon Dioxide (CO<sub>2</sub>)

Atmospheric CO<sub>2</sub> concentrations are projected to also increase under projected climate change scenarios. Since 1972, atmospheric CO<sub>2</sub> concentrations have increased from 324 ppm to 414 ppm, with an average increase of 2.3 ppm per year (Lindsey, 2020). All plant species are highly dependent on atmospheric CO<sub>2</sub> for photosynthesis. There are three main types of photosynthesis: C<sub>3</sub>, C<sub>4</sub>, and crassulacean acid metabolism (CAM). Elevated CO<sub>2</sub> will stimulate C<sub>3</sub> photosynthesis by reducing the loss of CO<sub>2</sub> through photorespiration and increasing the concentration gradient of CO<sub>2</sub> from air to leaf (Barnaby and Ziska, 2012). C<sub>4</sub> plants concentrate CO<sub>2</sub> around Rubisco, therefore reducing photorespiration, thus increasing CO<sub>2</sub> will not affect the photosynthetic rate of C<sub>4</sub> plants (Barnaby and Ziska, 2012). Corn utilizes the C<sub>4</sub> photosynthetic pathway. CAM plants are similar to C<sub>4</sub> plants although their stomates are closed during the day and open at night unlike C<sub>4</sub> plants (Kumar et al., 2017).

While it is suggested that increasing CO<sub>2</sub> will benefit C<sub>3</sub> plants over C<sub>4</sub> plants for the reasons outlined above, a phenomenon called photosynthesis down-regulation may occur negating these benefits. Photosynthetic down-regulation can be defined as a sustained, reversible

decline in photosynthetic activity associated with unfavorable environmental conditions (Gamon et al., 2001). Previous research conducted on soybean (C<sub>3</sub>) and corn (C<sub>4</sub>) reported that when exposed to CO<sub>2</sub> at 3000 and 5000 ppm, both species experienced a photosynthetic down-regulation. However, Kim et al. (2007) found that when corn was exposed to enhanced CO<sub>2</sub> concentrations of 750 ppm net photosynthesis increased an average of 10%, but was accompanied by lower stomatal conductance and transpiration compared to plants grown under 370 μmol mol<sup>-1</sup>.

Aside from photosynthesis, increased CO<sub>2</sub> concentrations may also impact plant growth and reproduction. Rudorff et al. (1996) reported that elevated CO<sub>2</sub> concentrations did not impact corn plant total biomass, total grain weight, and 1000-grain weight, but slightly decreased corn seed weight. Kim et al. (2007) reported that elevated CO<sub>2</sub> did not impact corn leaf area or aboveground biomass accumulation of the leaves, stalks, or ears.

Overall, weed species are genetically more diverse compared to crops, therefore it is possible weeds may show growth and reproductive responses to elevated CO<sub>2</sub> (Amare, 2016). Ziska et al. (1999) reported a significant increase in net photosynthesis and decrease in stomatal conductance of common lambsquarters (C<sub>3</sub>), under elevated CO<sub>2</sub>, but no impact on redroot pigweed (*Amaranthus retroflexus* L.) (C<sub>4</sub>) photosynthetic rates. In general, how species will be impacted by enhanced CO<sub>2</sub> concentrations over a long period of time is still unclear (Amare, 2016).

One way to measure photosynthetic efficiency is by using a new instrument called MultispeQ. MultispeQ, designed by a collaborative group at Michigan State University, was developed to create a high-throughput scientific instrument that was inexpensive, user friendly, adaptable to many environments, and capable of sharing data across the PhotosynQ platform.

Using the MultispeQ instrument in the field allows researchers to be able to measure a wide range of environmental and phenotypic parameters in response to abiotic stressors such as drought (Kuhlgert et al., 2016). Previous research has shown photosynthetic efficiency of non-drought tolerant cowpea (*Vigna unguiculate* (L). Walp., C<sub>3</sub>) genotypes to be statistically reduced ( $P \leq 0.05$ ) when under drought stress (Mwale et al., 2017). Mwale et al. (2017) also reported a strong correlation ( $r = 0.75$ ,  $P \leq 0.001$ ) between chlorophyll content and photosynthetic efficiency on all non-drought tolerant genotypes, implying that those parameters might be useful in identifying drought tolerance. Additional research on cowpea has shown a 64.6% reduction in photosynthesis under drought stress (Cardona-Ayala et al., 2020). Another method that can be used to identify drought tolerance is to identify a quantitative trait loci (QTL) molecular marker that associates with drought tolerance (Yu et al., 2019). In turfgrass, identifying and selecting grass varieties that are drought tolerant can be difficult, but recent studies have shown that visual ratings of leaf wilting can also be a reliable approach to identifying drought tolerance in the field without going through molecular processes (Yu et al., 2019).

### **Drought and Corn**

Future climate change scenarios projected precipitation will become more erratic with the majority of the total rainfall occurring in May and June and less during the growing season when it is most critical (Dai et al., 2016). More specifically in the Great Lakes Region since 1951 there has been an 14% increase in total precipitation and a 35% increase in extreme precipitation events (GLISAP, 2017). While CO<sub>2</sub> concentrations and temperatures are relatively easy to project, precipitation patterns, intensities, and distributions are more difficult and therefore uncertain (Varanasi et al., 2016).

Genetic improvements in grain corn have resulted in hybrids with greater tolerance to many abiotic and biotic stresses (Tollenaar and Lee, 2002). Water deficiency is a critical abiotic stress to corn (Witt et al., 2012). Water deficiency can greatly impact corn growth and development. Moisture stress during the elongation phase of corn leaves can slow down plant growth which can ultimately lead to reductions in yield (Boyer, 1970; Denmead and Shaw, 1960), although in general corn plants seem to be more tolerant to moisture stress during vegetative growth stages than reproductive (Claassen and Shaw, 1970). Substantial yield decreases can occur if drought events overlap with the reproductive growth stages of silking and grain fill (Claassen and Shaw, 1970). For example, if drought stress occurs 7-10 days before silking, delayed silk development can occur (Licht and Archontoulis, 2017). In addition, drought stress during grain fill can result in premature death of leaves, fewer kernels, and low kernel weights (Licht and Archontoulis, 2017).

In general, under drought conditions in which water availability is low and temperatures are high, stomates will close to reduce the loss of water through transpiration. During transpiration, water and oxygen are moving out of the stomates and CO<sub>2</sub> is moving in. When transpiration is stopped or slowed down, leaves will wilt, and photosynthesis rate and efficiency will slow down. Photosynthetic efficiency rates have been found to decrease during times of lower water potential or moisture stress in corn and soybean when grown in a growth chamber (Boyer, 1970).

Water use efficiency (WUE) is considered a main determinant factor of crop yield and drought resistance (Blum, 2009). With the current climatic conditions, crop breeders began to develop crops with higher WUE in order to help combat the negative results of drought stress (O'Shaughnessy et al., 2019). However, crop's photosynthetic pathway (C<sub>3</sub>, C<sub>4</sub>, or CAM) will

play a large role in the crop's WUE (Knee and McMahon, 2011). Plants that possess the C<sub>4</sub> or CAM photosynthetic pathway have an advantage over C<sub>3</sub> plants because they are able to still maintain high photosynthetic efficiency under drought stress, due to their ability to concentrate CO<sub>2</sub> around Rubisco and bundle sheath cells (Leakey et al., 2019).

To combat the stress that corn hybrids experience during drought conditions, many agricultural seed companies have developed corn hybrids that are more tolerant to stress induced by drought conditions. DroughtGard™ corn was developed by Bayer Crop Science through the constitutive expression of cold shock protein B (cspB) from *Bacillus subtilis* to improve performance of corn under drought conditions (Wang et al., 2015). cspBs from the bacterial species *B. subtilis* bind to single-stranded nucleic acids, therefore acting as a RNA chaperone (Zeeb and Balbach, 2003). Functionally, cspB enables corn plants to decrease the rate of water absorption from the soil in dry conditions, therefore enabling the corn plant to withstand drought conditions for longer periods of time than conventional non-drought tolerant hybrids (Eisenstein, 2013).

### **Drought and Weeds**

Moisture stress plays a critical role in weed physiology, growth, and development. With expected changes in precipitation, weed communities are likely to become more complex (Singh et al., 2011). Weed communities are likely to become more complex under projected climate change scenarios because they will invade new areas due to their strong response to changing climate compared to native species (Jinger et al., 2017). Non-native weed species are more likely to respond to the projected climate change scenarios because within the given environment, native species are less likely to adapt in the new temperature levels, CO<sub>2</sub> concentrations, or precipitation patterns (Hobbs and Mooney, 2005). These changes will also impact weed

management in agronomic fields because herbicide programs currently used for that specific weed community may not include control of the new invasive species (Hobbs and Mooney, 2005). It is projected that geographical migrations, weed species diversity, competition, and interactions with crops are going to shift under projected moisture stress (Ramesh et al., 2017). However, how species respond to drought is dependent on the species itself and cropping scenario (Amare, 2016). For example, if drought conditions were to occur for an extended period of time during spring germination, weed communities would shift to favor more deep-rooted species, whereas as early emerging species, which are often shallow rooted, will become suppressed (Sheley et al., 1996). Drought conditions can also favor weed seed survival in the soil as dry soil conditions do not favor many seed predators (Korres et al., 2016).

Weed species WUE will impact community composition and diversity under climate change projections. During a drought, the first defense mechanism is stomatal closure, this process is much quicker than other physical and physiological changes (Atteya, 2003). Species that are more drought tolerant highly regulate stomatal closure, thus improving their WUE (Atteya, 2003). Atteya (2003) reported that there was a 55% reduction in photosynthetic efficiency in drought stressed corn plants at tasseling, compared to vegetative growth stages. Weeds that possess the C<sub>4</sub> photosynthetic pathway are suggested to have a higher WUE than C<sub>3</sub>, resulting in C<sub>4</sub> weeds having the greatest advantage under hot dry conditions. In contrast C<sub>3</sub> weeds will outcompete C<sub>4</sub> weeds in saturated soils (Singh et al., 2011). Regardless of what photosynthetic pathway a weed possesses, Patterson (1986) found that when drought conditions are coupled with elevated CO<sub>2</sub> concentrations, the elevated CO<sub>2</sub> concentrations may reduce the effects of the drought, however the effects of enriched CO<sub>2</sub> are still expected to be greater in C<sub>3</sub> weeds.

## Drought and Crop - Weed Competition

When crops and weeds are grown in the same environment, crop yield can significantly be reduced due to competition. Previous research has shown that, crop-weed interactions will vary depending on geographic region and altered climatic conditions such as temperature, precipitation, and soil type (Singh et al., 2011). The type of photosynthetic pathway a plant utilizes plays a large role in the outcome of crop-weed competition. However, in a cropping system, the crop is usually competing with a combination of C<sub>3</sub> and C<sub>4</sub> weed species (Singh et al., 2011).

In a greenhouse study, Wiese and Vandiver (1970) evaluated the growth and competitive ability of sorghum (*Sorghum bicolor* (L.) Moench ssp. *bicolor*., C<sub>4</sub>), corn (C<sub>4</sub>), and eight weed species at different soil moisture levels. They reported that regardless of soil moisture and weed competition, corn produced the most biomass. Furthermore, kochia (*Bassia scoparia* (L.) A. J. Scott., C<sub>4</sub>) and Russian thistle (*Salsola tragus* L., C<sub>4</sub>), were found to be most competitive under low soil moistures. In contrast, barnyardgrass (*Enchinochloa crus-galli* (L.) P. Beauv., C<sub>4</sub>) and common cocklebur (*Xanthium stumarium* (L.)., C<sub>3</sub>) were found to be most competitive under high soil moistures. Overall, in a survey evaluating crop competition with weeds where water stress was the main variable evaluated and results demonstrate there was a slight tendency for low water availability to favor the crop, thus reducing the competitive impact of the weed Patterson (1995a).

Drought impacts on plants include decreased growth rate and photosynthesis (Mwanamwenge et al., 1999). In a greenhouse study evaluating the photosynthetic efficiency rates of soybean (C<sub>3</sub>) and velvetleaf (C<sub>3</sub>) under varying water stress treatments, Munger (1987) reported under low water stress, velvetleaf photosynthetic efficiency was approximately two

times greater than soybean, but under high water stress velvetleaf photosynthetic efficiency decreased rapidly and soybean decreased at a steady rate. Results from this study suggest that in drought conditions, velvetleaf will be less competitive with crops.

### **Drought and Weed Control**

The plant cuticle is a layer of wax and cutin that are on the outer surface of the leaf. One function of the cuticle is to restrict transpiration thus increasing tolerance to drought conditions (Goodwin and Jenks, 2005). The thickness of the waxy layer within the cuticle and the amount of pubescence also plays a critical role in herbicide absorption (Patterson, 1995b). Adjuvants are additives to herbicide spray solutions that aid in the absorption of herbicides through the waxy layer of the leaf cuticle. Zollinger (2012) conducted a study comparing adjuvant effectiveness with glyphosate on grass and broadleaf control under drought conditions. Results demonstrate that high rates of surfactants need to be used on drought stressed weeds for adequate control (Zollinger, 2012). Zollinger (2012) also reported that out of all adjuvants tested, methylated seed oil (MSO) resulted in the greatest control compared to non-ionic surfactant (NIS) and crop oil concentrate (COC) when applied to drought stressed weeds (Zollinger, 2012).

Furthermore, photosynthesis rates are lower in drought stressed plants, leading to reductions in sugar production and subsequent translocation of herbicides (Parker and Boydston, 2005). In a recent study Skelton et al. (2016) reported that under drought stress, glyphosate absorption and translocation were reduced in waterhemp, but 2,4-D absorption and translocation were not altered. Boydston et al. (1992) reported that when green foxtail plants were drought stressed control from 2-4 D was reduced by 40%, compared to non-drought stressed plants. Boydston et al. (1992) also reported that absorption and translocation of  $^{14}\text{C}$ -fluzafop-P was similar between drought stressed and non-drought stressed green foxtail. The researchers

concluded that there were no differences in absorption and translocation of <sup>14</sup>C-fluzaiifop-P between drought stressed and non-drought stressed green foxtail because, herbicide application may not alter the plants' response if the drought stress is terminated shortly after application (Boydston, 1992). Furthermore, even when extended 4 and 10 days after treatment, no difference was detected, thus it is possible that the specific SOA of <sup>14</sup>C-fluzaiifop-P is less sensitive to inhibition by the herbicide on drought stressed plants (Boydston, 1992).

In addition to drought impacting specific SOA herbicide absorption and translocation of foliar applied herbicides, reductions in soil moisture also play a role in preemergence herbicide efficacy (Skelton et al., 2016). Preemergence herbicides are herbicides applied to the soil prior to plant emergence. For preemergence herbicides to be effective they must be incorporated into the top 50 mm of soil by rain or mechanical methods to control newly germinating weed seeds. A study conducted by Moraes et al. (2018) found that when sulfentrazone was applied preemergence during drought conditions, greater than 80% control of morning glory species occurred, but when imazapic was applied during drought conditions, only 60% of morning glory species were controlled. Additionally, imazapic and sulfentrazone effectiveness began to decrease after 30 and 60 days of drought (Moraes Ribeiro et al., 2018). Under non-drought conditions, imazapic provided roughly 79-97% control on morning glory species, whereas sulfentrazone provided roughly 95-100% control (Moraes Ribeiro et al., 2018). Microbial degradation may also play an important role in residual herbicide efficacy (Moraes Ribeiro et al., 2018).

In conclusion, with future climate change projections, it is important that producers have an action plan to help mitigate potential negative impacts on crop growth, weed competition, and yield. With the introduction of drought tolerant corn hybrids, it is still unclear how these hybrids

will respond to extensive drought conditions, as well as different weed pressures. It is also important to know how weed communities in the Great Lakes Region will respond to drought conditions. Therefore, the objectives of these studies were to evaluate the performance of drought tolerant corn hybrids under drought stress with varying weed pressures and assess how water stress and crop competition impact weed density, growth, and community composition.

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## LITERATURE CITED

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## CHAPTER II

### ASSESSING WATER STRESS ON CORN HYBRID PERFORMANCE AND WEED COMMUNITIES

#### Abstract

Water stress and weed competition are critical stressors during corn (*Zea mays* L.) development. Genetic improvements in corn have resulted in hybrids with greater abiotic stress tolerance; however, drought remains problematic. Therefore, with the expected change in precipitation throughout the Great Lakes Region, field studies were conducted in East Lansing, Michigan (2019-2020) to evaluate weed competition and water stress on drought tolerant corn performance and weed community composition. The study followed a completely randomized design with four replications. Factorial combinations consisted of hybrid (drought tolerant, DT or drought sensitive, DS), weed pressure (weed free, 50%, or no control), and precipitation (ambient or 70% reduced). Corn growth and development was measured at four growth stages. Weed species density was measured three times and biomass collected. At harvest, corn ears were harvested for yield component analysis. Data were analyzed in R using linear mixed effects, diversity indexes, and non-linear regression models. Weed density was not modified by corn hybrid. However, in July weed density was lower under reduced than ambient precipitation ( $p = 0.001$ ). Furthermore, weed communities under reduced precipitation were more diverse in July than weed communities under ambient precipitation ( $p = 0.02$ ). Species evenness was more uniform under reduced precipitation in July ( $p = 0.009$ ). There was a significant main effect of weed pressure ( $p < 0.0001$ ) and precipitation ( $p = 0.007$ ) for corn yield. Averaged across weed pressures and precipitation, corn yield was not different between DT and DS hybrids ( $p = 0.893$ ). Overall, corn yield was reduced by 31% under 100% weed competition compared to weed free or 50%. Results demonstrate that reduced precipitation and increasing weed pressure decreases

corn yield and impacts species diversity and evenness. Ultimately, integrated weed management will need to adapt to these changes for continued success under future climate scenarios.

## Introduction

Projected climate change scenarios will have many negative effects on Michigan and Great Lakes Region's agricultural production. Since 1951, the Great Lakes Region has seen an increase in annual average air temperature of  $-16.5^{\circ}\text{C}$ , 14% increase in total precipitation, and a 35% increase in heavy precipitation events (GLISAP, 2017). Additionally, when those precipitation events occur, the Great Lakes Integrated Sciences and Assessments program hypothesizes that they will become more erratic and intense, resulting in many days during the growing season with little or no rainfall. Considering those projections, it is important to understand how drought conditions might impact corn production and weed communities in the Great Lakes Region.

Water stress is a critical abiotic stress during corn development (Witt et al., 2012). While genetic improvements in grain corn have resulted in many hybrids with greater tolerance to abiotic and biotic stressors, it is clear that drought stress is still problematic (Tollenaar and Lee, 2002). Given that, recent focus on genetic improvements in corn have shifted to drought tolerance in order to obtain high yields even during the drought years (Campos et al., 2004).

Since the commercialization of genetically engineered drought tolerant corn in 2012, adoption within the United States increased exponentially with approximately 22% or 7.5 million hectares planted with drought tolerant corn in 2016 compared to 2% in 2012 (McFadden et al., 2019). Specifically, in Michigan, 20% of the corn acreage planted in 2016 were drought tolerant hybrids (McFadden et al., 2019). DroughtGard™ corn was developed by Bayer Crop Science through the constitutive expression of cold shock protein B (cspB) from *Bacillus subtilis* to improve performance of corn under drought conditions (Wang et al., 2015). cspBs from the bacterial species *B. subtilis* bind to single-stranded nucleic acids, therefore acting as a RNA

chaperone (Zeeb and Balbach, 2003). Functionally, *cspB* enables corn plants to decrease the rate of water absorption from the soil in dry conditions, therefore enabling the plant to withstand drought conditions for longer periods of time than conventional non-drought tolerant hybrids (Eisenstein, 2013).

Previous research has documented mixed results on the efficacy of drought tolerant (DT) corn hybrids. Roth et al. (2013) in Indiana found there was no differences in grain yield between DT and drought sensitive (DS) corn hybrids. They also reported that photosynthesis and transpiration rates at the leaf-scale were not statistically significant between DT and DS hybrids. Furthermore, in a field study including six different experiments at four locations in Kansas with varying evapotranspiration environments, DT hybrids had higher water use efficiency (WUE) than DS hybrids under high (>508 mm) and medium (432 to 488 mm) evapotranspiration conditions but were similar under low (< 432 mm) evapotranspiration conditions (Adee et al., 2016). In conjunction with higher WUE in DT hybrids under high and medium evapotranspiration environments, there was also a 5-7% yield increase (Adee et al., 2016).

Another yield limiting factor in grain corn production is weed competition. Since the beginning of agriculture, weeds have competed with desired crops for light, water, and nutrients (Boehm et al., 2011). Weed communities are a group of weed species that localize within the same habitat at the same time (Booth et al., 2003). With the projected changes in climate, it is suggested that weed communities are likely to become more complex with the capability to invade new areas (Jinger et al., 2017). Non-native weed species are more likely to respond positively to projected climate change scenarios. Native species are less likely to adapt to the new temperature levels, CO<sub>2</sub> concentrations, or precipitation patterns because these disturbances disrupt the structure of the native ecosystem (Hobbs and Mooney, 2005). These changes will also

impact weed management in agronomic fields because herbicide programs currently used for the specific weed community, may not control new species. In addition, with projected geographical migrations, weed species diversity, competition, and interactions with crops will shift under projected moisture stress (Ramesh et al., 2017). However, how weed species respond to drought conditions is dependent on the species itself and the cropping scenario (Amare, 2016). Under the projected climate change, weed species WUE will impact community composition and diversity. It is hypothesized that during extended drought conditions, C<sub>4</sub> weeds will outcompete C<sub>3</sub> weeds (Rodenburg et al., 2010). Reasons for this shift surround photosynthetic efficiency, C<sub>4</sub> weeds have a higher WUE than C<sub>3</sub> weeds, thus C<sub>4</sub> weeds will be more dominant under dry environmental conditions (Singh et al., 2011).

Climate change impacts resulting in irregular and intense rainfall are already impacting the Great Lakes Region (Patz et al., 2008). In 2016, 20% of corn acres were planted with DT corn hybrids in Michigan; however, with future climate scenarios, it is expected that this percentage will increase (McFadden et al., 2019). In addition, with only 15% of the total farmland in Michigan is irrigated, DT hybrids may help farmers maintain yield during drought conditions without having to invest in costly irrigation equipment (Kisekka et al., 2015; USDA-NASS, 2017). With limited research on DT hybrid performance in rainfed Michigan systems and the project increase in erratic rainfall events, Michigan grain corn producers must be aware of how these hybrids perform under reduced precipitation and added weed pressure. Therefore, the objectives of this research were to: 1) determine how reduced precipitation and crop competition impact weed density, growth, and community composition and 2) compare performance of DT and DS corn hybrids under reduced precipitation and varying weed pressure.

## Materials and Methods

Field experiments were conducted in 2019 at the Michigan State University Crop and Soil Sciences Research Farm in East Lansing, MI (42°42'34.55"N, 84°27'56.15"W) and in 2020 at the Michigan State University Plant Pathology Farm in East Lansing, MI (42°41'19.30"N, 84°29'21.83"W). The soil type at the Crop and Soil Science Research Farm was a sandy loam Aubbeenaubbee-Capac soil with a pH of 6 and 2.1% organic matter with 18 cm<sup>3</sup>/cm<sup>3</sup> plant available water (NRCS, 2020). The soil type at the Plant Pathology Farm was a clay loam Colwood-Brookston soil with a pH of 6.8 and 2.7% organic matter with 27 cm<sup>3</sup>/cm<sup>3</sup> plant available water (NRCS, 2020). The soil was prepared by fall chisel plow followed by two passes in the spring with a soil finisher. Previous crop for both experimental years was soybean. A preplant incorporated urea fertilizer (46-0-0) was applied at 168 kg ha<sup>-1</sup> followed by 112 L ha<sup>-1</sup> of 16-16-16 (N-P-K) applied at planting 5 cm down and 5 cm over from the seed.

Two isoline corn hybrids, DKC47-27 DroughtGard<sup>®</sup> Double Pro (drought tolerant, DT) and DKC46-20 Triple Pro (drought sensitive, DS) (Bayer Crop Science, St. Louis, MO, USA) were planted using a four row Almaco packet planter with 76 cm row spacing at a population of 85,000 seeds ha<sup>-1</sup>. Corn was planted on May 17, 2019 and May 25, 2020. Plots were 3 m wide (4 rows) and 7 m long. A preemergence herbicide application of Acuron (bicyclopyrone + mesotrione + atrazine + S-metolachlor) (Syngenta, Wilmington, DE, USA) was made after planting on May 17, 2019 using a tractor-mounted, compressed air sprayer and May 26, 2020 using a backpack sprayer calibrated to deliver 178 L ha<sup>-1</sup> at 206 kPa using AIXR 11003 nozzles (TeeJet Technologies, Wheaton, IL). Two rates of the herbicide were applied to achieve desired weed pressures: 160 L ha<sup>-1</sup> and 53 L ha<sup>-1</sup>.

The experimental design followed a three-factor randomized complete block design with four replications. Factorial combinations consisted of hybrid (DT or DS), weed pressure (weed free, 50% weeds, or 100% weeds), and precipitation (ambient or reduced). Rainout shelters were constructed out of PVC and clear corrugated polycarbonate panels following the methods of Yahdjian and Sala (2002). Rainout shelters were 3 m long and 3 m wide and were placed over four corn rows in the middle of the plot. Panels were placed on the top of the shelter and constructed to cover 70% of the total area, with the goal of reducing total ambient precipitation by 70%; however, only 30-50% of the total ambient precipitation was calculated to be collected. Gutters were installed on the front of the shelters to eliminate rain pooling. Buffer strips of four corn rows were used to eliminate rain from the gutters moving into adjacent plots. Two (2019) and one (2020) 1 m<sup>2</sup> sub-plots were installed in each plot for subsequent weed species evaluation prior to weed emergence.

Precipitation data was obtained throughout the growing season using rain gauges. One gauge was placed above the corn canopy and captured total ambient rainfall and one gauge was placed below the corn canopy to measure total ambient rainfall that penetrated the soil surface through the corn canopy. To measure how much rainfall the shelters intercepted, on a subset of shelters randomly distributed amongst replications, a hose was connected to the gutter and precipitation drained into a collection bin. To ensure that the shelters were not altering the microclimate beneath, iButton's were installed on a subset of plots to evaluate the temperature and relative humidity. Soil moisture was measured throughout the growing season using a TDR Field Scout in two locations for subplot at 12 cm in depth.

Corn measurements consisted of height, stem diameter, growth stage, photosynthetic output, and anthesis-silking interval (ASI). Measurements were taken at four significant growth

stages known to be impacted by water stress: V6, V12, VT/R1, R3/R4 (Cakir, 2004; Ritchie et al., 1997). Ten (2019) and five (2020) plants were tagged in the center two rows in each plot to track plant development throughout the season. Corn height was measured from the ground level to the base of the highest leaf collar. Stem diameter was measured at the base of the plant using a digital caliper. Growth stage was measured during the vegetative growth stage by counting leaf collars and then during the reproductive growth stage by evaluating kernel development.

Photosynthesis readings (Phi2, quantum yield of photosynthesis II; PhiNO, quantum yield of other unregulated losses; and PhiNPQ, quantum yield of non-photochemical quenching) were taken using a MultispeQ V 2.0 (PhotosynQ Inc., East Lansing, MI, USA). The reading took place on five tagged corn plants on the middle leaf. Once corn reached R1, photosynthesis readings were taken on the ear leaf. Photosynthesis measurements were taken from 8:00 AM to 12:00 PM on clear sunny days. Anthesis-silking interval (ASI) was evaluated for each tagged plant daily in late July for anther and silk emergence (Lindsey and Thomison, 2016).

Additionally, ASI was calculated by subtracting the date of silk emergence minus the date of anther emergence. Corn growing degree day (GDD) was calculated using 86/50 cutoff.

Corn ears were hand harvested after natural plant senescence on October 15, 2019 and October 19, 2020 and placed in a dryer for 2 d at 66°C. At harvest, the main ears of the tagged plants were hand-harvested, shelled, and evaluated for moisture on a plot basis. The following yield components were measured: rows per ear, kernels per row, total kernel count, and weight from individually shelled ears (Zamaninejad et al., 2013).

Weed measurements consisted of density counts by species. Weed measurements were taken three times during the growing season (June, July, and at harvest). At harvest, weeds were

collected by individual species and placed in a dryer for 3 d at 66°C. After plants were dried, species were weighed for total biomass.

### Statistical Analysis

Field experiment data were combined over years after examining side-by-side box plots of the residuals and conducting Levene's test for unequal variances. Furthermore, normality assumptions were assessed by examining normal probability plots of the residuals and histograms. Corn growth parameters, yield estimate parameters, weed density, and soil volumetric water content (VWC) were analyzed using the lmer function from the lme4 package in R (R, 2021). Hybrid, weed competition, and precipitation were considered fixed effects, and replication, year, and replication nested within year were considered random effects. Differences in means were further investigated using Tukey's HSD post hoc test in the emmeans package in R (R, 2021). Additionally, corn growth parameters were analyzed using the drc package in R (R, 2021) following the methods outlined in Knezevic et al. (2007). Three parameter log-logistic models were fit to corn height, growth stage, and stem diameter (equation 1) using the drc modelFit function.

$$Y = \frac{d}{1 + \exp [b(\log x - e)]} \quad [1]$$

For equation 1,  $Y$  is the response variable (height, growth stage, or stem diameter),  $x$  is the total accumulated GDD,  $d$  is the upper limit,  $b$  is the relative slope around  $e$ , and  $e$  is the inflection point (Streibig, 1988). Models were used to predict the GDD's needed to reduce relative corn height, growth stage, and stem diameter by 25 and 50%. Percent reduction data are GDD's relative to the control of no weed competition, ambient precipitation, and drought sensitive corn hybrid. Anthesis-silking intervals (ASI) were analyzed using logistic regression to assess the influence that weed competition, precipitation, and corn hybrid had on occurrence of ASI's

considered extreme (+/-2) (Edmeades et al., 2000). Weed species diversity was assessed using Simpson's Diversity Index (equation 2) and species evenness (equation 4).

$$D = \frac{\sum [n_i(n_i-1)]}{[N(N-1)]} \quad [2]$$

Simpson's Diversity Index was used to quantify the biodiversity within each weed community (Nkoa et al., 2015). For Simpson's Diversity Index,  $D$  represents diversity,  $\Sigma$  is the sum of all the factors that follow,  $n_i$  is the density or number of the  $i$ th species, and  $N$  is the total number of individuals of all the species in the community.

$$H' = \Sigma[-p_i(\ln p_i)] \quad [3]$$

The Shannon-Wiener Diversity Index was used to derive species evenness.  $H'$  represented the diversity index,  $\Sigma$  is the sum of all the factors that follow, and  $p_i$  is the proportional abundance or relative frequency of the  $i$ th species (equation 3).

$$E = \frac{H'}{\ln S} \quad [4]$$

Species evenness was used to determine if a weed community was dominated by one or more species or whether the species were evenly distributed throughout the community (Booth et al., 2010).  $E$  represented species evenness,  $H'$  is the Shannon-Wiener Diversity Index, and  $S$  is the species richness (i.e., total number of species).

## **Results and Discussion**

### **Precipitation and Soil Moisture**

In 2019, there was 10.3 cm more precipitation than in 2020 (Table 2.1). Overall, averaged across hybrids and levels of weed competition, reduced precipitation treatment decreased VWC by 21% throughout the season ( $p = <0.0001$ ). During the months of May, July, August, September, and October there was a significant main effect of precipitation ( $p = <0.001$ ). Averaged across levels of weed competition and hybrids, reduced precipitation

decreased VWC content by 23, 28, 29, 27, and 18% in May, July, August, September, and October, respectively (Table 2.2). During the month of June, there was significant three-way interaction between hybrid, weed competition, and precipitation ( $p = 0.04$ ), where reduced precipitation decreased VWC when DT hybrids were present for each level of weed competition by 16, 15, and 16% compared to DT hybrids under ambient precipitation (Table 2.2,  $p = 0.0001$ ,  $0.002$ , and  $0.0001$ ). Furthermore, in June treatments with DS hybrids under reduced precipitation decreased VWC in weed free and 50% weed competition treatments by 24 and 16%, respectively, compared to DS hybrids grown under ambient precipitation (Table 2.2,  $p = <0.0001$  and  $0.0001$ ). Additionally, DT hybrids under reduced precipitation and increasing weed competition from weed free to 50 and 100% competition decreased soil moisture by 19 and 16% compared to DT hybrids under ambient precipitation and no weed competition (Table 2.2,  $p = <0.0001$  and  $0.0002$ ). In October, there was a significant main effect of precipitation and weed competition (Table 2.2,  $p = <0.0001$  and  $0.03$ ). Specifically, during October, averaged across hybrids and weed competition treatments, reduced precipitation decreased VWC by 18% compared to ambient precipitation (Table 2.2). Additionally, averaged across hybrids and precipitation treatments, VWC increased by 8% in the 50% weed competition treatment compared to a weed free treatment (Table 2.2,  $p = 0.02$ ).

Overall, the precipitation patterns observed in 2019 and 2020, partially agree with the projections by Dai et al. (2016) where they projected that there will be more heavy rainfall events in the months of May and June followed by fewer events during July and August. In 2019 and 2020, the majority of the precipitation occurred at the beginning of the growing season during vegetative growth of corn with less during the middle of the growing season coinciding with the beginning reproductive growth stages (Table 2.1). Water stress during vegetative and

reproductive growth stages can negatively influence corn growth and development (Witt et al., 2012). Specifically, at the end of the vegetative growth stage and the beginning of the reproductive growth stage, water stress can negatively influence pollination and kernel number, resulting in a reduction in yield (Lee, 2007; Ritchie et al., 1997).

### Weed Community Composition

Weed density was not modified by corn hybrid or precipitation in June ( $p = 0.34$  and  $0.07$ ). However, there was a significant main effect of weed competition level on weed density, averaged across corn hybrids and precipitation treatments weed density under 50% weed competition was 88% lower than 100% weed competition treatment (Figure 2.1a,  $p = <0.0001$ ). Additionally, in June, averaged across precipitation treatments, weed communities grown in competition with DS hybrids were less diverse than weed communities grown in competition with DT hybrids (Figure 2.2a,  $p = 0.02$ ). Furthermore, in June, averaged across precipitation treatments, weed communities grown in competition with DS hybrids were less even than weed communities grown in competition with DT hybrids (Figure 2.2b,  $p = 0.008$ ).

Weed density was not modified by corn hybrid in July ( $p = 0.08$ ). However, there was a significant main effect of level of weed competition and precipitation on weed density in July ( $p = <0.0001$  and  $<0.0001$ ). Averaged across hybrids and precipitation treatments, weed density of the 50% weed competition treatment was 64% lower compared to the 100% weed competition treatment (Figure 2.1b,  $p = <0.0001$ ). Additionally, averaged across hybrids and weed competition treatments, weed density was 44% lower under reduced precipitation compared to ambient precipitation (Figure 2.1b,  $p = <0.0001$ ). Furthermore, weed communities under reduced precipitation were more diverse and even in July than weed communities under ambient precipitation (Figure 2.2c-d,  $p = 0.01$  and  $0.007$ ).

To our knowledge, this is the first experiment conducted in the Midwest that evaluated the influence of reduced precipitation on weed density and diversity in competition with corn hybrids with differing levels of drought tolerance. However, studies evaluating drought impacts on weed species diversity have been conducted in the Mediterranean shrubland in Israel and Eastern Mediterranean Basin (Alon and Sternberg, 2019; Tielbörger et al., 2014). Alon and Sternberg (2019) reported that drought conditions significantly increased weed species diversity and composition. However, after a nine-year study evaluating long-term drought effects, Tielbörger et al. (2014) reported no significant changes in species diversity or composition, however other researchers suggest that no differences in community composition were detected due to the reduction in precipitation only being 30% (Knapp et al., 2017; Smith, 2011). Furthermore, our experiment demonstrated similar results to Alon and Sternberg (2019) where reduced precipitation increased weed species diversity and evenness. However, we believe that the reduced precipitation treatment only affected species diversity later in the growing season due to the large amount of precipitation that occurred at the beginning of the growing season, resulting in little resource competition to occur between the species due to water not being a limiting resource. Additionally, we speculate that weed species diversity and evenness increased under drought conditions later in the growing season due to drought removing or reducing weed density therefore allowing other species to invade vacated gaps within the community. We observed this with species such as common lambsquarters (*Chenopodium album* L.) and eastern black nightshade (*Solanum ptychanthum* Dunal) increasing in density under reduced precipitation compared to ambient precipitation treatments (data not shown).

## Corn

### *Height*

Reduced precipitation, increasing weed competition, and corn hybrids with differing levels of drought tolerance did not negatively affect the number of GDD's required to reach a 25% reduction in corn height (Table 2.3). Additionally, holding weed competition level and corn hybrid constant, reduced precipitation did not affect the number of GDD's required to reach a 50% reduction in corn height (Table 2.3). However, the number of GDD's required to reach a 50% reduction in corn height when DS corn hybrid were grown with no weed competition under reduced precipitation was 15, 20, and 18 more GDD's than DT corn hybrids under 50% and 100% weed competition and ambient precipitation and DS corn hybrids grown under 100% weed competition and ambient precipitation (Table 2.3,  $p = 0.05, 0.01, \text{ and } 0.04$ ).

Holding weed competition and precipitation constant, the DT corn hybrid did not influence the number of GDD's required to reach a 50% reduction in corn height compared to the DS corn hybrid (Table 2.3). Overall, increasing weed competition and reduced precipitation had little influence on height of corn hybrids with differing levels of drought tolerance. In contrast, Licht and Archontoulis (2017) reported water stress during the vegetative growth stages reduce stem and leaf cell expansion, resulting in shorter plants. However, Ge et al. (2012) reported that in a field experiment, differences in corn height among three water stress treatments (33, 55, and 75%) were not different 21 days after treatment.

### *Stem Diameter*

Holding weed competition and corn hybrid constant, reduced precipitation did not increase the number of GDD's required to reach a 25 or 50% reduction in corn stem diameter (Table 2.3). However, DT hybrids grown under 50% weed competition and under reduced

precipitation reached a 25% reduction in stem diameter 28 GDD's faster than DS hybrids grown without weed competition and under ambient precipitation (Table 2.3,  $p = 0.04$ ).

Furthermore, DS hybrids grown with no weed competition and ambient precipitation reached a 50% reduction in stem diameter 27, 41, and 46 GDD's slower than DT hybrids grown under weed free, 50%, or 100% weed competition and reduced precipitation, respectively (Table 2.3,  $p = 0.04$ , 0.003, and 0.001). Additionally, DS hybrids grown without weed competition and under ambient precipitation reached a 50% reduction in stem diameter 38 and 50 GDD's slower than DS hybrids grown under 100% weed competition and with either ambient or reduced precipitation (Table 2.3,  $p = 0.03$  and 0.006).

Overall, these results demonstrate that there were few differences in the number of GDD's required to reach a 25% reduction in corn stem diameter amongst treatments in our study (Table 2.3). Additionally, holding corn hybrid constant and weed competition constant, reduced precipitation did not decrease the number of GDD's required to reach a 50% reduction in corn stem diameter compared to ambient precipitation (Table 2.3). These results differ from previous research by Ge et al. (2012) where they reported that an increase in water stress, decreases stem diameter, regardless of plant developmental stage. However, adding the DT trait and reducing precipitation decreased the number of days required to reach a 50% reduction in corn stem diameter compared to hybrids with the DS trait under no weed competition and ambient precipitation (Table 2.3). Furthermore, under greenhouse conditions Butts et al. (2017) reported that weed presence did not decrease corn stem diameter compared to no weed presence until the V8 and V11 corn growth stages.

### *Growth Stage*

Holding corn hybrid and weed competition constant, decreasing precipitation did not decrease the number of GDD's required to reach a 25 or 50% reduction in corn growth stage (Table 2.3). Furthermore, DT hybrids grown under 50% weed competition and ambient precipitation reached a 25% reduction in corn growth stage 24 GDD's faster than DT hybrids grown under 100% weed competition and reduced precipitation (Table 2.3,  $p = 0.02$ ). Under ambient precipitation, DT corn grown under 50% weed competition reached a 25% reduction in corn growth stage 22 GDD's faster than DS corn grown under 100% weed competition and ambient precipitation (Table 2.3,  $p = 0.04$ ).

Furthermore, DT hybrids grown under ambient precipitation and 50% weed competition reached a 50% reduction in growth stage 34 and 50 GDD's faster than DT hybrids grown under 100% weed competition and either ambient or reduced precipitation (Table 2.3,  $p = 0.02$  and  $0.0005$ ). Additionally, DS hybrids grown under weed free or 50% weed competition and either ambient or reduced precipitation reached a 50% reduction in growth stage 43, 39, 45, and 38 days faster than DT hybrids grown under 100% weed competition and reduced precipitation (Table 2.3,  $p = 0.005, 0.01, 0.004, \text{ and } 0.02$ ) Overall, the phenology of corn hybrids with differing levels of drought tolerance is regulated by level of weed competition and the amount of precipitation.

### *Anthesis-Silking Interval*

Averaged across hybrids, a two-way interaction occurred between weed competition and precipitation for the ASI (Figure 2.3,  $p = 0.005$ ). Under no weed competition and 100% weed competition, reduced precipitation did not increase or decrease ASI (Figure 2.3,  $p = 0.98$ ). However, under 50% weed competition and ambient precipitation the ASI was 0.69 days more

negative compared to reduced precipitation (Figure 2.3,  $p = <0.0001$ ). Furthermore, increasing weed competition from no weed competition under reduced precipitation to 50% weed competition under ambient precipitation negatively decreased the ASI by 0.55 days (Figure 2.3,  $p = 0.004$ ). Additionally, increasing weed competition from no weed competition under ambient precipitation to 50% weed competition under reduced precipitation the ASI was increased by 0.439 days (Figure 2.3,  $p = 0.01$ ). Furthermore, increasing weed competition from 50% weed competition under ambient precipitation to 100% weed competition under ambient or reduced precipitation increased the ASI by 1.20 and 1.29 days (Figure 2.3,  $p = <0.0001$  and  $<0.0001$ ). Additionally, increasing weed competition from 50% weed competition under reduced precipitation to 100% weed competition under ambient or reduced precipitation increased the ASI by 0.51 and 0.61 days (Figure 2.3,  $p = 0.002$  and  $0.0001$ ). Previous research conducted on ASI in corn reported intervals increasing up to 30 days (Edmeades et al., 2000).

ASI data were further analyzed using logistic regression to assess the influence that weed competition, precipitation, and corn hybrid had on occurrence of ASI's considered extreme ( $\pm 2$ ) (Edmeades et al., 2000). Positive ASI's occur when the corn silks emerge before the anthers and a negative ASI occurs when the anthers emerge before the silks. The odds of corn hybrids having an ASI greater than or equal to positive two was 2.79 times more likely for corn grown under 100% weed competition than no competition (Table 2.4,  $p = 0.08$ ). Furthermore, for each  $g^{-ear}$  increase in yield, the odds of corn hybrids having an ASI greater than or equal to positive two decrease by 3% (Table 2.4,  $p = <0.0001$ ). However, the odds of corn hybrids having an ASI greater than or equal to positive two was not significant when comparing ambient versus reduced precipitation ( $p = 0.89$ ) or DS versus DT hybrids ( $p = 0.96$ ) (Table 2.4). Furthermore, the odds of corn hybrids having an ASI less than or equal to negative two was 6.35 times more likely for

corn grown under 50% weed competition compared to 100% weed competition ( $p = 0.004$ ) and 4.75 times more likely for corn grown under 100% weed competition than no competition ( $p = 0.02$ ) (Table 2.4). Additionally, the odds of DS hybrid having an ASI less than or equal to negative two is 0.58 more likely than the DT hybrid (Table 2.4,  $p = 0.08$ ).

Overall, our results demonstrate that it is more likely to have extreme ASI's when corn has no weed control, regardless of precipitation treatment. Further, Reid et al. (2014) reported that under weed free conditions DT hybrids was found to have a shorter ASI compared to DS hybrids, although weed competition lengthened the ASI for both hybrids (Reid et al., 2014). ASI's that are extreme can influence corn pollination, ultimately reducing corn yield. When corn is exposed to a biotic or abiotic stress during the pollination stages, the interval between pollen shedding and silk emergence can be increased or decreased, resulting in a positive or negative ASI (Edmeades et al., 2000).

### *Photosynthesis*

Hybrid, precipitation, or weed competition did not reduce Phi2 levels when measured at V4-V6 (Table 2.5). However, a significant two-way interaction occurred between hybrid and weed competition when Phi2 was measured at R3-R4 (Table 2.5,  $p = 0.008$ ). Holding corn hybrid and weed competition constant, reduced precipitation did not increase Phi2 levels for DT or DS hybrids (Table 2.5). However, averaged across precipitation treatments, Phi2 levels in DT hybrids with no weed competition were 12% higher compared to DT hybrids with 100% weed competition and 12% higher than DS hybrids without weed competition (Table 2.5). Phi2, or quantum yield of photosystem II photochemistry, is the percentage of incoming light that goes into photosystem II, which is a measure of photosynthetic efficiency (Kramer et al., 2004). From these results we can conclude that photosynthetic efficiency is not modified by reduced

precipitation, weed competition, or drought tolerant status during early vegetative growth. However, when DT hybrids are grown under extreme weed competition during reproductive growth photosynthetic efficiency is reduced. Furthermore, photosynthetic efficiency is reduced in DS hybrids without weed competition compared to DT hybrids without weed competition during reproductive growth. Previous research has also concluded that drought stress significantly reduced Phi2 levels in cowpea (Mwale et al., 2017).

Hybrid, precipitation, or weed competition did not increase PhiNPQ levels when measured at V4-V6 (Table 2.5). When PhiNPQ was measured at R3-R4, there was a significant three-way interaction between corn hybrid, precipitation, and weed competition (Table 2.5,  $p = 0.07$ ). Holding weed competition and corn hybrid constant, reducing precipitation did not increase PhiNPQ levels at the R3-R4 growth stage (Table 2.5). However, DT hybrids grown under 100% weed competition and reduced precipitation had PhiNPQ levels 31% higher than DT hybrids grown without weed competition and ambient precipitation (Table 2.5,  $p = 0.002$ ). Additionally, DS hybrids grown under no weed competition and reduced precipitation had PhiNPQ levels 33% higher than DT hybrids grown under no weed competition and ambient precipitation (Table 2.5,  $p = 0.08$ ). Furthermore, DT hybrids grown under 100% weed competition and reduced precipitation had PhiNPQ levels 30% higher than DS hybrids grown under 100% weed competition and reduced precipitation (Table 2.5,  $p = 0.07$ ).

PhiNPQ, or quantum yield of non-photochemical energy, measures energy loss via down regulation of photochemistry (Kramer et al., 2004). From these results we can conclude that PhiNPQ levels are not modified by reduced precipitation, weed competition, or DT hybrids during early vegetative growth. A similar response occurred with Phi2 levels. Additionally, we can conclude that reduced precipitation does not increase PhiNPQ levels at the R3-R4 growth

stage, however, increasing weed competition modifies that interaction by increasing PhiNPQ levels. Additionally, presence of the DT trait under weed competition and reduced precipitation has higher PhiNPQ levels than the DS trait under reduced precipitation and no weed competition. This interaction suggests that hybrids that contain the DT trait will have higher PhiNPQ levels under weed competition and drought stress, which results in energy loss via down regulation of photochemistry.

Hybrid, precipitation, or weed competition did not decrease PhiNO levels when measured at V4-V6 (Table 2.5). When PhiNO was measured at R3-R4, there was a significant three-way interaction between corn hybrid, precipitation, and weed competition (Table 2.5,  $p = 0.01$ ). Holding corn hybrid and weed competition constant, reduced precipitation did not decrease PhiNO levels in any treatments except for DT hybrids grown under 100% weed competition (Table 2.5). Specifically, DT hybrids grown with 100% weed competition and reduced precipitation had 14% lower PhiNO levels than DT hybrids grown with the same level of weed competition and ambient precipitation (Table 2.5,  $p = 0.05$ ). Additionally, DS hybrids grown under 50% weed competition and ambient precipitation had 16% higher PhiNO levels than DT hybrids grown under 100% weed competition and reduced precipitation (Table 2.5,  $p = 0.05$ ).

PhiNO is the quantum yield of other unregulated processes (Kramer et al., 2004). From these results we can conclude that the quantum yield of other unregulated processes is not modified by reduced precipitation, weed competition, or drought trait status during early vegetative growth stage. A similar response occurred with Phi2 and PhiNPQ levels. These results also demonstrate that DT hybrids under reduced precipitation decrease PhiNO levels under 100% weed competition. Therefore, DT hybrids under reduced precipitation and weed competition

have the likelihood of having higher photosynthetic efficiency due to PhiNO unregulated processes not inhibiting photosynthesis or being harmful to the plant.

Overall the PhotosynQ instrument allows for the evaluation of sensitive indicators of various photosynthetic parameters and the ability to view the onset of photoinhibition and photodamage that may be caused by plant stress (Baker and Rosenqvist, 2004). To our knowledge, there has not been a study conducted evaluating photosynthetic parameters on drought stressed drought tolerant corn and weed competition using the PhotosynQ instrument. However, our results demonstrate that early in the vegetative development of corn, reduced precipitation, weed competition, and the presence of a DT trait do not modify the photosynthetic parameters evaluated in this study. However, during the R3-R4 growth stage, reducing precipitation and increasing weed competition modifies photosynthetic processes. Previous research has been conducted on the photosynthetic response of drought stressed drought tolerant corn hybrids using a Li-COR meter. A greenhouse study conducted by Wijewardana et al. (2017) reported that photosynthetic rates measured five times during the study declined in DT corn hybrids as soil moisture levels decreased. Therefore, due to these evaluations only occurring twice throughout the growing season, future research should target other critical growth stages where weed competition and drought stress are known to influence corn growth.

#### *Yield Components*

When evaluating corn yield ( $\text{g}^{-\text{ear}}$ ), a significant three-way interaction occurred between hybrid, weed competition, and precipitation (Figure 2.4,  $p = 0.05$ ). Under no weed competition, there was no difference between DT and DS hybrids under ambient or reduced precipitation (Figure 2.4,  $p = 1.0$  and  $1.0$ ). However, holding corn hybrid constant under no weed competition, reducing precipitation decreased corn yield for DT and DS hybrids by 14 and 15% (Figure 2.4,  $p$

= 0.004 and 0.005). Additionally, under 50% weed competition, there was no difference between DT and DS hybrid under ambient precipitation ( $p = 1.0$ ); however, under reduced precipitation, DT hybrids had 20% lower yield than DS hybrids ( $p = 0.006$ ) (Figure 2.4). Furthermore, holding corn hybrid constant under 50% weed competition, reducing precipitation decreased corn yield for DT and DS hybrids by 31 and 14% (Figure 2.4,  $p = <0.0001$  and 0.007). Additionally, under 100% weed competition, there was no difference between DT and DS hybrids under ambient or reduced precipitation (Figure 2.4,  $p = 0.89$  and 0.58). Holding corn hybrid constant under 100% weed competition, reducing precipitation decreased corn yield for the DT hybrid by 23% ( $p = 0.003$ ); however, reduced precipitation did not decrease corn yield for the DS hybrid ( $p = 1.0$ ) (Figure 2.4). Furthermore, with a DT hybrid, increasing weed competition from no weed competition under ambient precipitation to 100% weed competition under ambient or reduced precipitation, decreased corn yield by 24 and 42% (Figure 2.4,  $p = <0.0001$  and  $<0.0001$ ). These responses correspond to the ASI results in which we demonstrated that by increasing weed competition from no competition to 100% competition, the odds the ASI will be extreme ( $> 2$  or  $< -2$ ) is 2.79 and 4.75 times more likely, thus having detrimental yield impacts (Table 2.4).

We further wanted to explore the effect of incremental increases in weed density on the rate of corn yield loss (Figure 2.5). As weed density increased there was no difference in the rate of yield loss between DS hybrids under ambient or reduced precipitation (Figure 2.5,  $p = 0.41$ ). In contrast, as weed density increased the rate of yield loss was 1.8 times greater for DT hybrids grown under ambient precipitation compared to DT hybrids grown under reduced precipitation (Figure 2.5,  $p = 0.006$ ). Furthermore, as weed density increased the rate of yield loss was 1.3 times greater for DS hybrids grown under ambient precipitation compared to DT hybrids grown under reduced precipitation (Figure 2.5,  $p = 0.01$ ). There was no difference in the rate of yield

loss as weed competition increased between DT and DS hybrids grown under ambient precipitation ( $p = 0.78$ ) and DT and DS hybrids grown under reduced precipitation ( $p = 0.09$ ) (Figure 2.5).

Overall, corn hybrids with differing levels of drought tolerance differed in their ability to buffer the detrimental impacts of increasing weed competition on yield. Therefore, we further wanted to explore what component of yield: row number per ear, kernels per row, and (or) total kernel count were driving these differences. There was no difference in the rate of row number per ear loss between any treatment combinations investigated in this study (Figure 2.6).

Although reduced precipitation did not influence the rate of row number per ear loss for DT hybrids as weed density increased, kernels per row was affected. As weed density increased, the rate of kernels per row loss between DT hybrids grown under ambient precipitation was 1.6 times faster compared to DT hybrids grown under reduced precipitation (Figure 2.7,  $p = 0.04$ ). However, reduced precipitation did not decrease the rate of kernels per row loss for DS hybrids compared to ambient precipitation (Figure 2.7,  $p = 0.35$ ). Furthermore, the rate of kernel number per row loss as weed density increased was not different between DT and DS hybrids grown under ambient precipitation ( $p = 0.93$ ) or reduced precipitation ( $p = 0.26$ ) (Figure 2.7). However, the rate of kernel per row loss for DS hybrids grown under ambient precipitation was 1.25 times faster compared to DT hybrids grown under reduced precipitation (Figure 2.7,  $p = 0.05$ ). Additionally, the rate of kernel per row loss as weed density increased for DT hybrids grown under ambient precipitation was not different than DS hybrids grown under reduced precipitation (Figure 2.7,  $p = 0.30$ ).

Furthermore, corn hybrids with differing levels of drought tolerance responded to incremental increases in weed density in total kernel count (Figure 2.8). As weed density

increased, the rate of total kernel count loss between DS hybrids grown under ambient precipitation was 1.4 times faster compared to DT hybrids grown under reduced precipitation (Figure 2.8,  $p = 0.02$ ). In contrast, as weed density increased, there was no difference in the rate of total kernel count loss between DT hybrids grown under ambient precipitation and DS hybrids grown under reduced precipitation (Figure 2.8,  $p = 0.29$ ). Additionally, as weed density increased, there was no difference in the rate of total kernel count loss between DS hybrids grown under ambient or reduced precipitation (Figure 2.8,  $p = 0.23$ ). Interestingly, as weed density increased, the rate of total kernel count loss for DT hybrids grown under ambient precipitation was 1.5 times faster than DT hybrids grown under reduced precipitation (Figure 2.8,  $p = 0.02$ ). However, reduced precipitation did not decrease the rate of kernels count loss for DS hybrids compared to ambient precipitation (Figure 2.7,  $p = 0.35$ ). Furthermore, as weed density increased, the rate of total kernel loss was not different among hybrids grown under ambient ( $p = 0.88$ ) or among hybrids grown under reduced ( $p = 0.21$ ) precipitation (Figure 2.8).

Overall, our results demonstrate that DT hybrids buffer increasing levels of weed competition better when grown under reduced precipitation than when DT hybrids are grown under ambient precipitation. Furthermore, DT hybrids buffer increasing levels of weed competition better when grown under reduced precipitation than DS hybrids are grown under ambient precipitation. To our knowledge, no study has reported the influence that weed competition and reduced precipitation have on DT corn hybrid yield components. Previous research conducted on DT corn hybrids in the Northern Great Plains reported that there were no differences in grain yield between the DT hybrids and its isoline when grown under weed free conditions (Chang et al., 2014).

The projected change in climate for the Great Lakes Region will affect agriculture production in many ways. Since 1951, the Great Lakes Region has seen an increase in annual average air temperature of  $-16.5^{\circ}\text{C}$ , 14% increase in total precipitation, a 35% increase in heavy precipitation events (GLISAP, 2017). Given these projections, drought events may become more common in the Great Lakes Region. From these results, we can conclude that drought sensitive and drought tolerant hybrid performance is affected by reduced precipitation and the magnitude of these impacts are regulated by level of weed competition. Furthermore, during drought conditions, weed species communities will tend to become more diverse. An increase in diversity may result in difficulties with herbicide-based weed control, as well as the possibility that species that are normally not present within a system, could become more dominant. Therefore, future investigation should expand this research into multiple geographies to determine how DT hybrids will respond to different weed communities, soil conditions, and temperatures. Additionally, to further explore the influence that weed species communities could have on DT hybrid performance, researchers should evaluate herbicide and mechanical weed control programs to determine how they might respond to changes in weed community composition.

This study was the first step in identifying how climate will influence weed biology and drought tolerant corn production. Although genetic improvements to corn have been made resulting in corn with increased levels of drought tolerance it is evident weed competition under reduced soil moisture conditions will detrimentally impact yield. Therefore, taking proactive integrated weed management steps by identifying the corn-weed competition principles investigated in this study will allow field crop growers across the Great Lakes Region to integrate weed biology and crop physiology in the development of integrated economically viable weed management programs under future climate stress.

## **APPENDIX**

Table 2.1. Monthly precipitation at the study location in East Lansing, MI for 2019 and 2020.

	2019	2020	30-year average <sup>2</sup>
Month	Precipitation (mm)		
May	78.0 <sup>1</sup>	103.1 <sup>1</sup>	85.3
June	165.4	58.5	87.6
July	67.1	33.9	72.1
August	28.6	75.4	82.0
September	81.7	102.4	88.9
October	60.8	28.8	64.3
Total	481.6	402.1	480.3

<sup>1</sup>Precipitation data collected from rain gauges at both site years.

<sup>2</sup>Monthly 30-year average precipitation for Lansing, MI, data retrieved from NOAA Nation Centers for Environmental Information DOI:10.7289/V5PN93J

Table 2.2. Mean monthly (SE) soil volumetric water content (VWC) under two corn hybrids with differing levels of drought tolerance, three weed competition levels, and two precipitation levels in a two-year field experiment (2019-2020).

Treatment <sup>1</sup>			May* <sup>+</sup>	June	July* <sup>+</sup>	August* <sup>+</sup>	September* <sup>+</sup>	October*
Hybrid	WP	Precipitation	VWC (cm <sup>3</sup> /cm <sup>3</sup> )					
DT	W1	F	25.8 (1.1)b <sup>2</sup>	27.1 (1.3)ab <sup>3</sup>	16.1 (1.6)b <sup>2</sup>	7.4 (1.4)b <sup>2</sup>	13.4 (1.7)b <sup>2</sup>	20.3 (0.4)Ab <sup>2,4</sup>
DT	W1	R	20.2 (0.8)a	22.8 (1.5)cd	10.3 (1.4)a	5.4 (0.9)a	9.6 (1.5)a	16.4 (0.9)Aa
DT	W2	F	24.2 (1.5)b	25.9 (1.1)ab	13.6 (1.4)b	7.8 (1.6)b	13.8 (1.7)b	22.3 (0.6)Bb
DT	W2	R	20.0 (0.9)a	22.1 (1.4)cd	9.1 (1.3)a	5.2 (0.9)a	9.7 (1.5)a	18.2 (0.6)Ba
DT	W3	F	25.9 (0.8)b	27.2 (1.2)ab	13.0 (1.3)b	8.3 (1.6)b	13.7 (1.6)b	21.1 (0.5)ABb
DT	W3	R	20.7 (1.4)a	22.9 (1.3)cd	10.3 (1.1)a	6.1 (1.2)a	9.7 (1.7)a	17.7 (0.7)ABa
DS	W1	F	25.9 (1.5)b	28.2 (1.5)a	13.2 (1.7)b	7.0 (1.3)b	13.4 (1.7)b	21.4 (0.8)Ab
DS	W1	R	17.2 (1.6)a	21.5 (1.3)c	8.7 (1.3)a	5.0 (1.0)a	9.9 (1.4)a	17.1 (0.6)Aa
DS	W2	F	23.9 (0.9)b	27.1 (1.5)ab	15.3 (1.6)b	7.8 (1.5)b	13.6 (1.7)b	22.7 (0.4)Bb
DS	W2	R	18.5 (1.3)a	22.7 (1.5)cd	11.5 (1.4)a	5.6 (1.0)a	10.6 (1.6)a	18.2 (0.6)Ba
DS	W3	F	25.3 (1.1)b	24.9 (1.4)bd	12.7 (1.3)b	7.6 (1.5)b	13.4 (1.7)b	21.3 (0.5)ABb
DS	W3	R	19.2 (1.2)a	22.6 (1.4)cd	9.2 (1.2)a	5.3 (1.0)a	9.8 (1.3)a	18.3 (0.7)ABa

<sup>1</sup>Abbreviations: DT=drought tolerant corn hybrid, DS=drought sensitive corn hybrid, WP=weed competition, W1=weed free, W2=50% control, W3=no control, F=ambient precipitation, R=70% reduced precipitation.

<sup>2</sup>Means within the same column followed by the same lowercase letter are not significantly different ( $p \geq 0.05$ ) for the main effect of precipitation.

<sup>3</sup>Means within the same column followed by the same italicized capital letter are not significantly different ( $p \geq 0.05$ ) for the three-way interaction of hybrid, weed competition, and precipitation.

<sup>4</sup>Means within the same column followed by the same capital letter are not significantly different ( $p \geq 0.05$ ) for the main effect of weed competition.

\*Non-significant main effect of corn hybrid ( $p \geq 0.05$ ).

+Non-significant main effect of weed competition ( $p \geq 0.05$ ).

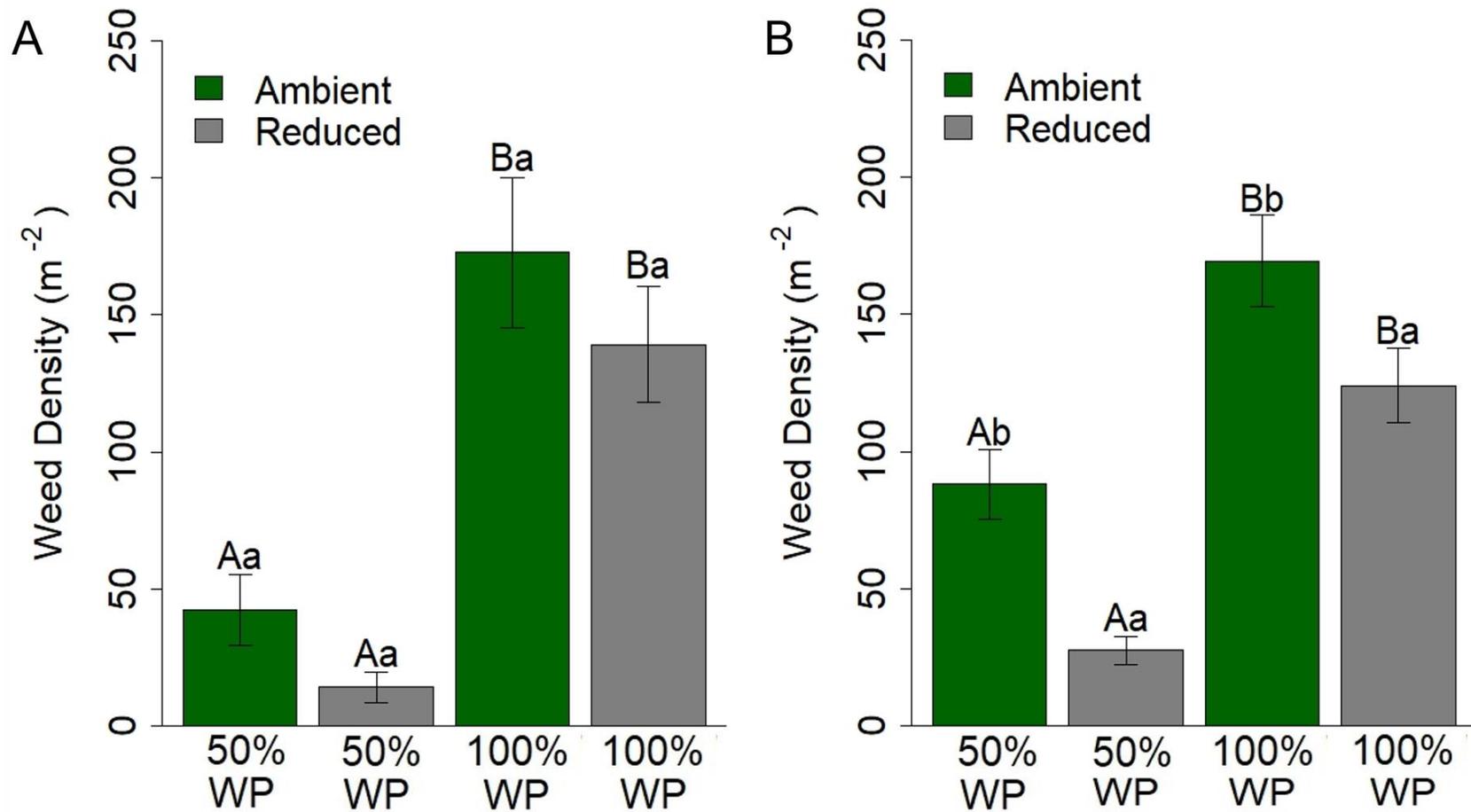


Figure 2.1. Mean weed density under two weed competition levels (50% WP, or 100% WP), two precipitation levels (ambient or 70% reduced), and averaged across two corn hybrids with differing levels of drought tolerance for the month of June (A) and July (B) in a two-year field experiment (2019-2020). Bars labeled by the same capital letter are not statistically different for main effect of weed competition ( $p \geq 0.05$ ). Bars labeled by the same lowercase letter are not statistically different for the main effect of precipitation ( $p \geq 0.05$ ).

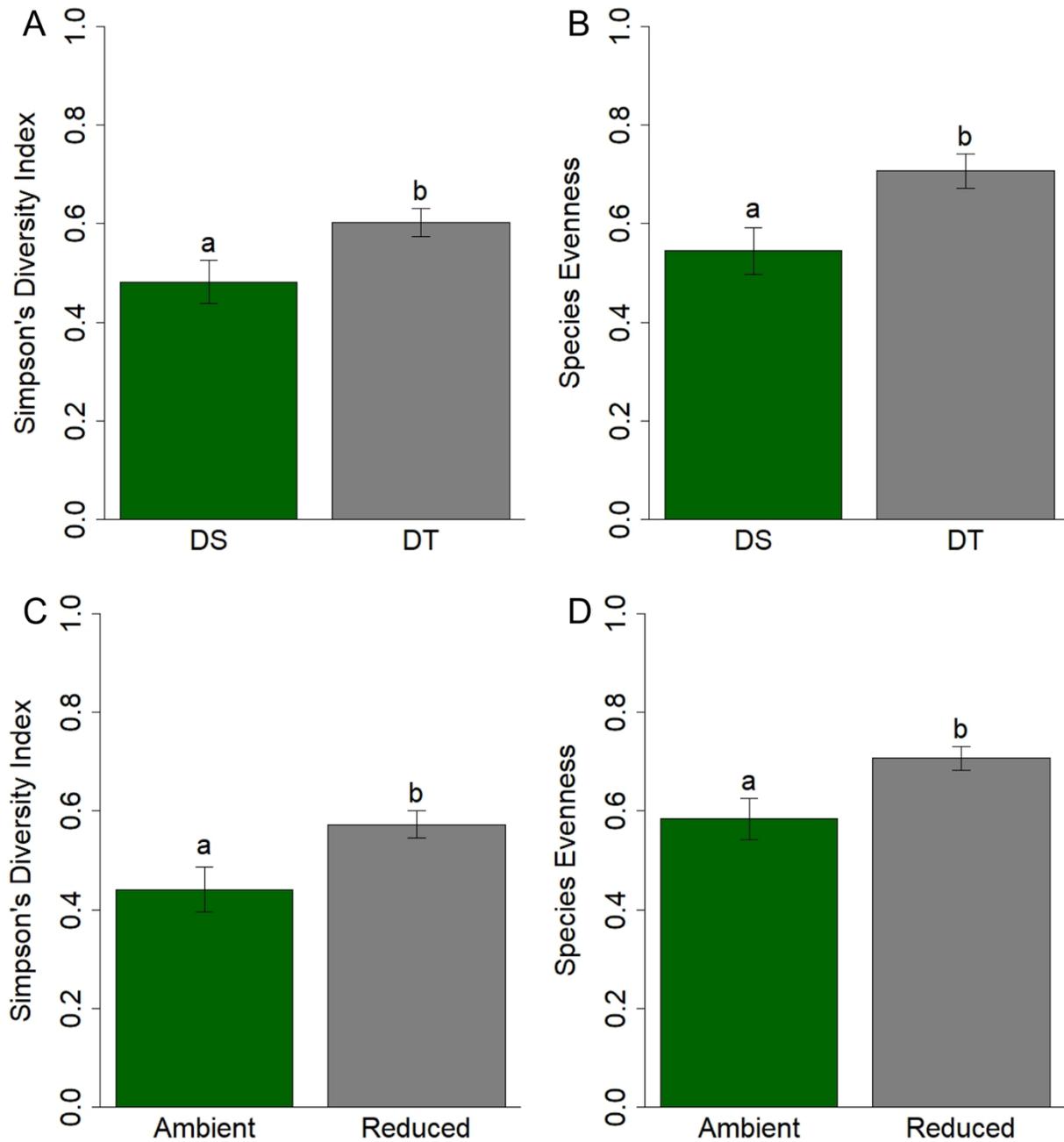


Figure 2.2. Weed community Simpson's Diversity Index and Species Evenness under two corn hybrids with differing levels of drought tolerance (DT=drought tolerant or DS=drought sensitive) and averaged across precipitation levels in June (A and B) or under two precipitation levels (ambient or 70% reduced) and averaged across corn hybrids in July (C and D) in a two-year field experiment (2019-2020). Bars labeled by the same lowercase letter are not statistically different ( $p \geq 0.05$ ).

Table 2.3. Mean (SE) accumulated growing degree days (GDD) required to reduce relative corn height, stem diameter, and growth stage by 25 and 50%, under two precipitation levels, two corn hybrids with differing levels of drought tolerance, and three weed competition levels in a two-year field experiment (2019-2020). Percent reductions and probability values were calculated by ED and EDcomp functions, respectively, in the drc package in R (R, 2021).

Treatments <sup>1</sup>			Relative height <sup>2</sup>		Relative stem diameter <sup>2</sup>		Relative growth stage <sup>2</sup>	
			25%	50%	25%	50%	25%	50%
Hybrid	WP	Precipitation	GDD					
DT	W1	F	331 (6.7)a <sup>3</sup>	388 (6.0)ab <sup>3</sup>	124 (9.9)ab <sup>3</sup>	192 (9.6)abc <sup>3</sup>	368 (11.3)abc <sup>3</sup>	667 (28.1)abc <sup>3</sup>
DT	W1	R	337 (7.1)a	398 (5.7)ab	115 (10.6)ab	177 (10.1)abd	366 (11.2)abc	665 (28.1)a
DT	W2	F	338 (7.4)a	388 (5.5)a	117 (9.4)ab	178 (8.9)abd	359 (11.2)a	659 (28.4)a
DT	W2	R	334 (7.0)a	392 (5.8)ab	102 (11.7)a	163 (11.3)ad	360 (11.5)ab	671 (29.4)abcd
DT	W3	F	331 (7.5)a	383 (6.2)a	115 (10.2)ab	174 (9.9)abd	373 (11.9)abc	693 (30.2)bcde
DT	W3	R	331 (7.8)a	387 (6.8)ab	107 (13.0)ab	158 (12.3)ad	383 (12.5)c	709 (30.8)e
DS	W1	F	338 (6.9)a	399 (5.6)ab	130 (9.6)b	204 (9.3)c	368 (11.2)abc	666 (28.0)abc
DS	W1	R	342 (6.9)a	403 (5.3)b	114 (10.5)ab	179 (10.0)abcd	365 (11.4)abc	670 (28.8)abcd
DS	W2	F	339 (7.1)a	393 (5.4)ab	128 (9.3)ab	195 (8.9)bc	366 (11.2)abc	664 (28.0)ab
DS	W2	R	338 (6.9)a	398 (5.5)ab	112 (10.5)ab	172 (9.8)abd	366 (11.4)abc	671 (28.8)abcd
DS	W3	F	332 (7.8)a	385 (6.6)a	104 (11.8)ab	166 (11.6)abd	381 (11.9)bc	699 (29.8)de
DS	W3	R	335 (7.4)a	398 (6.2)ab	101 (12.5)ab	154 (11.8)d	373 (11.9)abc	694 (30.2)cde

<sup>1</sup>Abbreviations: DT=drought tolerant corn hybrid; DS=drought sensitive corn hybrid; WP=weed competition; W1=weed free; W2=50% control; W3=no control; F=ambient precipitation; R=70% reduced precipitation.

<sup>2</sup>Percent reduction data are GDD's relative to the control of no weed competition, ambient precipitation, and drought sensitive corn hybrid.

<sup>3</sup>Means within the same column followed by the same lowercase letter are not significantly different for the three-way interaction of corn hybrid, weed competition, and precipitation level ( $p \geq 0.05$ ).

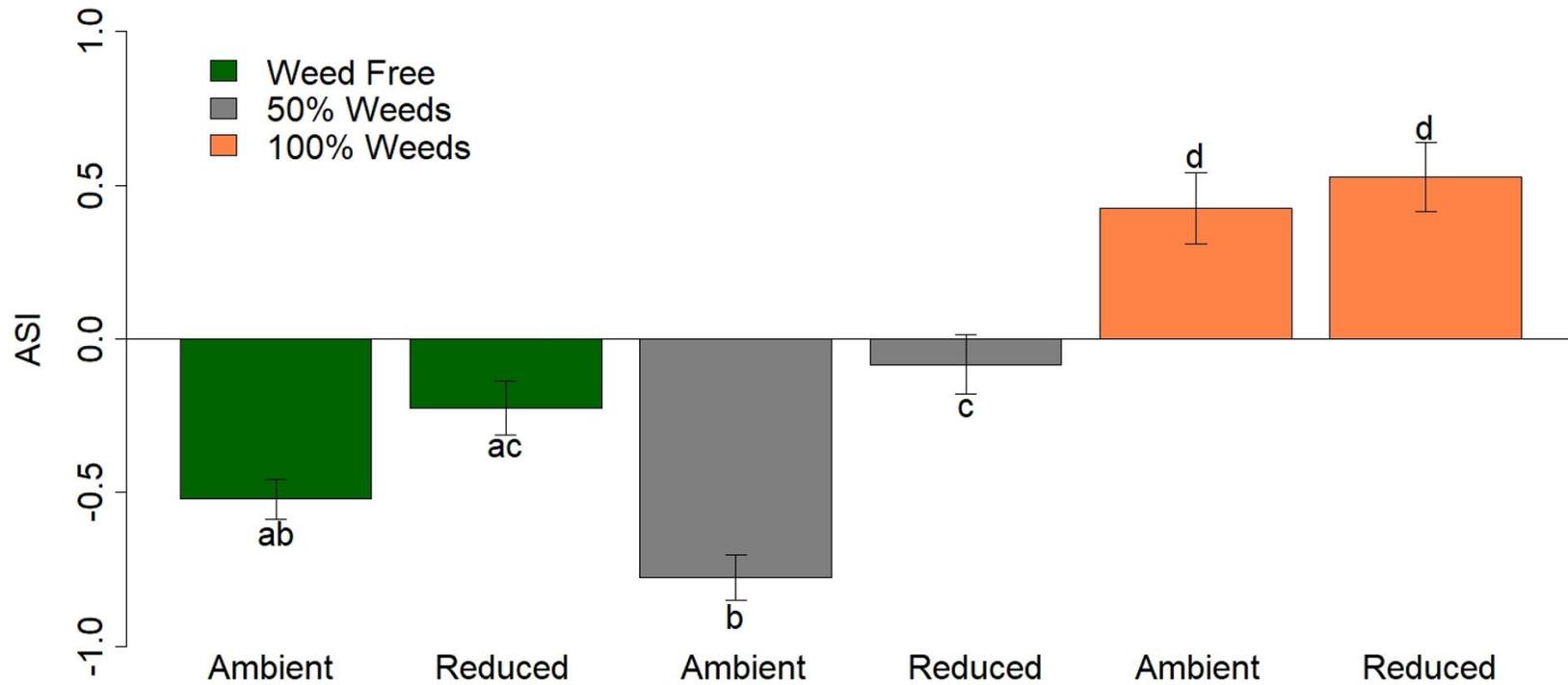


Figure 2.3. Mean (SE) anthesis-silking interval (ASI) in days under three weed competition levels (weed free, 50%, or 100%), two precipitation levels (ambient or 70% reduced), and averaged across two corn hybrids with differing levels of drought tolerance in a two-year field experiment (2019-2020). Bars labeled by the same lowercase letter are not statistically different ( $p \geq 0.05$ ).

Table 2.4. Summary of logistic regression analysis odds ratios of corn hybrids having an anthesis-silking interval (ASI) greater than or equal to positive two and less than or equal to negative two in a two-year field experiment (2019-2020).

Treatment comparisons <sup>1</sup>	Odds ratio <sup>2</sup>	
	Positive	Negative
F vs R	0.95	1.65
W1 vs W2	1.54	1.34
W2 vs W3	1.81	6.35**
W3 vs W1	2.79*	4.75**
DS vs DT	0.96	0.58*
Yield	0.97**	1.01**

<sup>1</sup>Abbreviations: DT=drought tolerant corn hybrid; DS=drought sensitive corn hybrid; WP=weed competition; W1=weed free; W2=50% control; W3=no control; F=ambient precipitation; R=70% reduced precipitation.

<sup>2</sup> Odds ratios followed by \* signifies  $p \leq 0.1$  and \*\* signify significance at  $p \leq 0.05$ .

Table 2.5. Mean (SE) corn photosynthetic response (Phi2, PhiNPQ, PhiNO) under three weed competition levels, two precipitation levels, and two corn hybrids with differing levels of drought tolerance at two corn growth stages in a two-year field experiment (2019-2020).

Treatments <sup>1</sup>			Phi2		PhiNPQ		PhiNO	
			Corn growth stage					
Hybrid	WP	Precipitation	V4-V6* <sup>+</sup>	R3-R4 <sup>+</sup>	V4-V6* <sup>+</sup>	R3-R4	V4-V6* <sup>+</sup>	R3-R4
DT	W1	F	0.48 (0.02)	0.53 (0.03) <i>a</i> <sup>3</sup>	0.36 (0.03)	0.27 (0.03) <i>a</i> <sup>2</sup>	0.16 (0.01)	0.20 (0.00) <i>ab</i> <sup>2</sup>
DT	W1	R	0.50 (0.01)	0.49 (0.03) <i>a</i>	0.34 (0.01)	0.32 (0.03) <i>abc</i>	0.16 (0.01)	0.19 (0.00) <i>ab</i>
DT	W2	F	0.49 (0.02)	0.46 (0.03) <i>ab</i>	0.35 (0.02)	0.34 (0.03) <i>abc</i>	0.16 (0.01)	0.20 (0.00) <i>ab</i>
DT	W2	R	0.48 (0.03)	0.47 (0.03) <i>ab</i>	0.35 (0.03)	0.34 (0.03) <i>abc</i>	0.17 (0.01)	0.19 (0.00) <i>ab</i>
DT	W3	F	0.46 (0.03)	0.47 (0.03) <i>b</i>	0.38 (0.04)	0.32 (0.03) <i>abc</i>	0.16 (0.01)	0.21 (0.00) <i>a</i>
DT	W3	R	0.49 (0.02)	0.43 (0.03) <i>b</i>	0.35 (0.01)	0.39 (0.04) <i>b</i>	0.17 (0.01)	0.18 (0.00) <i>b</i>
DS	W1	F	0.50 (0.02)	0.47 (0.03) <i>b</i>	0.34 (0.02)	0.33 (0.03) <i>abc</i>	0.17 (0.01)	0.20 (0.00) <i>ab</i>
DS	W1	R	0.47 (0.02)	0.46 (0.03) <i>b</i>	0.36 (0.03)	0.36 (0.03) <i>bc</i>	0.16 (0.01)	0.19 (0.00) <i>ab</i>
DS	W2	F	0.46 (0.02)	0.47 (0.03) <i>ab</i>	0.38 (0.02)	0.33 (0.03) <i>abc</i>	0.17 (0.01)	0.21 (0.00) <i>a</i>
DS	W2	R	0.47 (0.02)	0.47 (0.03) <i>ab</i>	0.37 (0.03)	0.33 (0.04) <i>abc</i>	0.16 (0.01)	0.20 (0.00) <i>ab</i>
DS	W3	F	0.47 (0.03)	0.46 (0.03) <i>ab</i>	0.36 (0.03)	0.34 (0.04) <i>abc</i>	0.17 (0.01)	0.19 (0.00) <i>ab</i>
DS	W3	R	0.48 (0.01)	0.50 (0.03) <i>ab</i>	0.35 (0.01)	0.30 (0.03) <i>ac</i>	0.18 (0.01)	0.20 (0.00) <i>ab</i>

<sup>1</sup>Abbreviations: DT=drought tolerant corn hybrid; DS=drought sensitive corn hybrid; WP=weed competition; W1=weed free; W2=50% control; W3=no control; F=ambient precipitation; R=70% reduced precipitation; Phi2=quantum yield of Photosystem II; PhiNPQ=quantum yield of non-photochemical quenching; PhiNO=quantum yield of other unregulated losses.

<sup>2</sup>Means within the same column followed by the same lowercase letter are not significantly different ( $p \geq 0.1$ ) for the three-way interaction of hybrid, weed competition, and precipitation.

<sup>3</sup>Means within the same column followed by the same italicized lowercase letter are not significantly different ( $p \geq 0.1$ ) for the two-way interaction of hybrid and weed competition.

\*Non-significant two-way interaction of corn hybrid and weed competition ( $p \geq 0.1$ ).

+Non-significant three-way interaction of corn hybrid, weed competition, and precipitation ( $p \geq 0.1$ ).

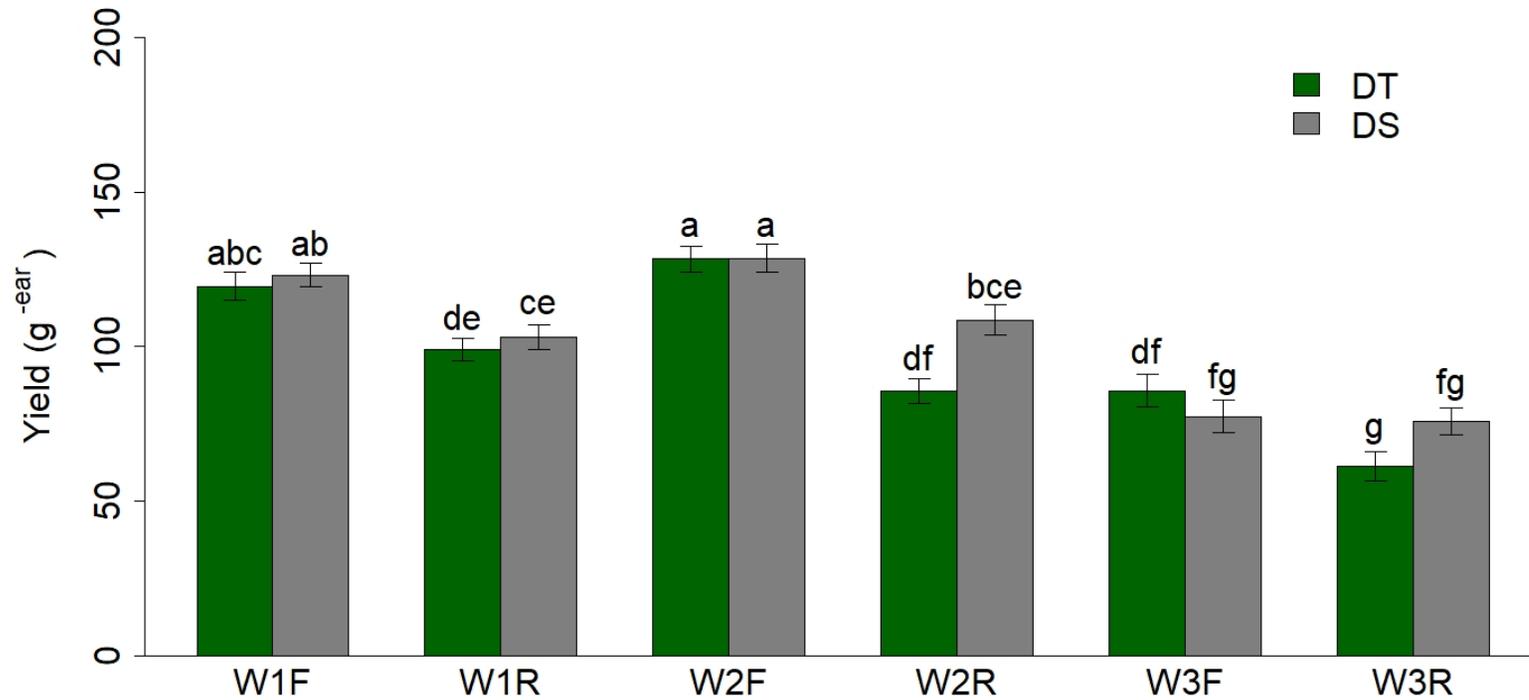


Figure 2.4. Mean (SE) yield (g<sup>-ear</sup>) under three weed competition levels (W1=weed free, W2=50%, or W3=100%), two precipitation levels (F=ambient or R=70% reduced), and two corn hybrids with differing levels of drought tolerance (DT=drought tolerant or DS=drought sensitive) in a two-year field experiment (2019-2020). Bars labeled by the same lowercase letter are not statistically different for the three-way interaction of corn hybrid, precipitation, and weed competition ( $p \geq 0.05$ ).

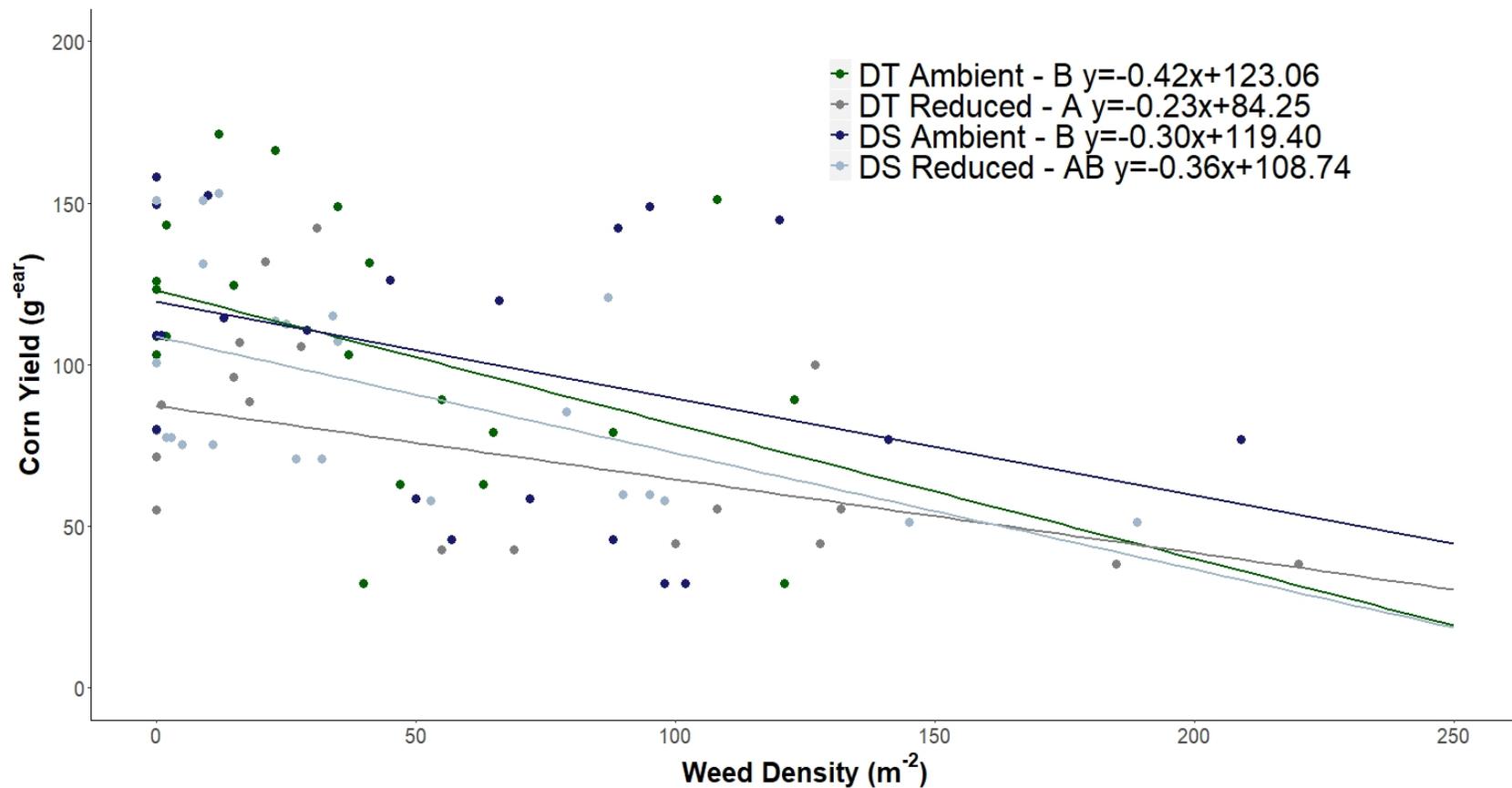


Figure 2.5. Corn yield (g<sup>-ear</sup>) as a function of weed density under two corn hybrids with differing levels of drought tolerance (DT=drought tolerant or DS=drought sensitive) and two precipitation levels (ambient or 70% reduced) in a two-year field experiment (2019-2020). Slopes obtained from regression lines labeled by the same capital letter are not statistically different ( $p \geq 0.05$ ).

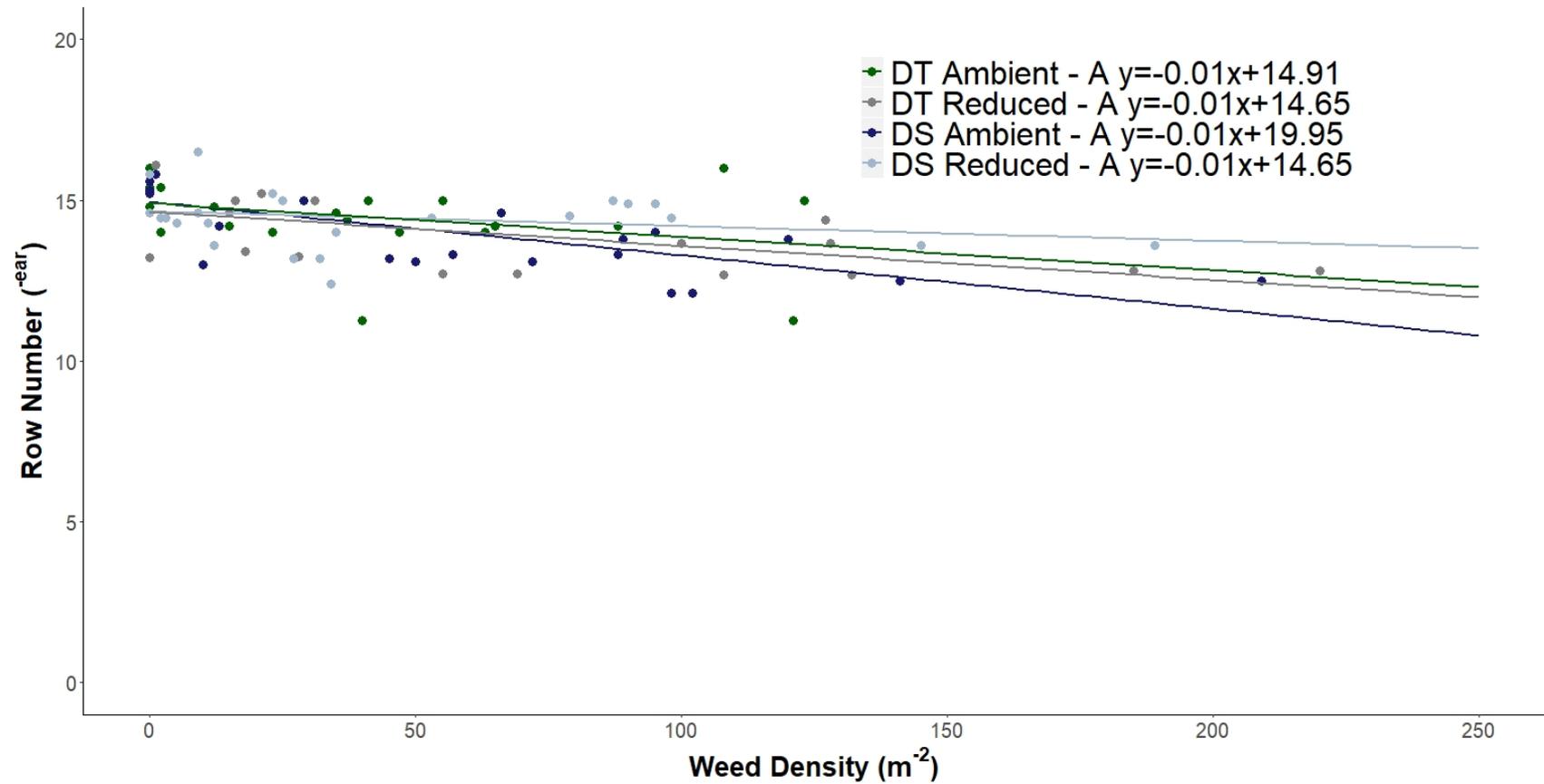


Figure 2.6. Corn row number (<sup>-ear</sup>) as a function of weed density under two corn hybrids with differing levels of drought tolerance (DT=drought tolerant or DS=drought sensitive) and two precipitation levels (ambient or 70% reduced) in a two-year field experiment (2019-2020). Slopes obtained from regression lines labeled by the same capital letter are not statistically different ( $p \geq 0.05$ ).

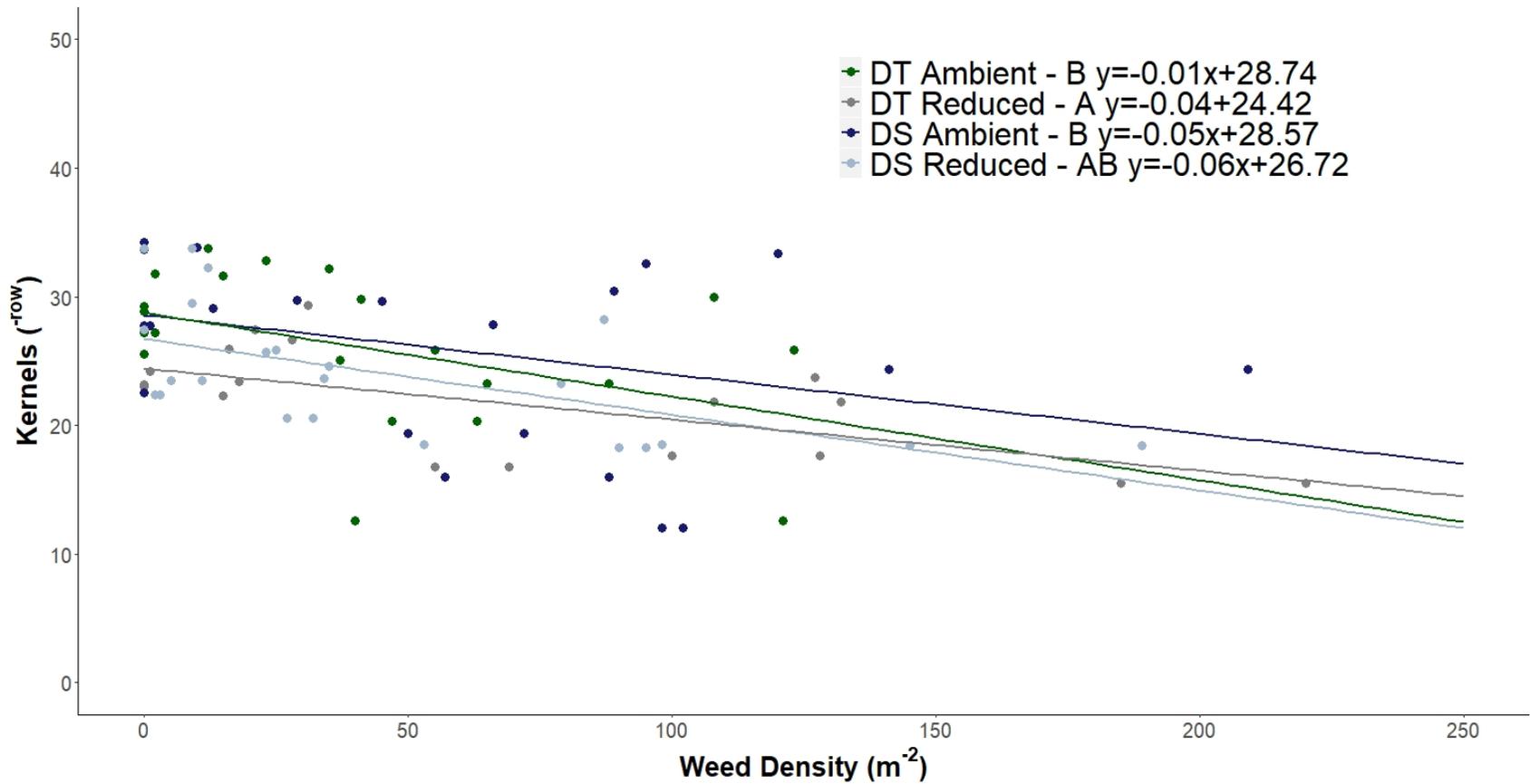


Figure 2.7. Corn kernels (<sup>-row</sup>) as a function of weed density under two corn hybrids with differing levels of drought tolerance (DT=drought tolerant or DS=drought sensitive) and two precipitation levels (ambient or 70% reduced) in a two-year field experiment (2019-2020). Slopes obtained from regression lines labeled by the same capital letter are not statistically different ( $p \geq 0.05$ ).

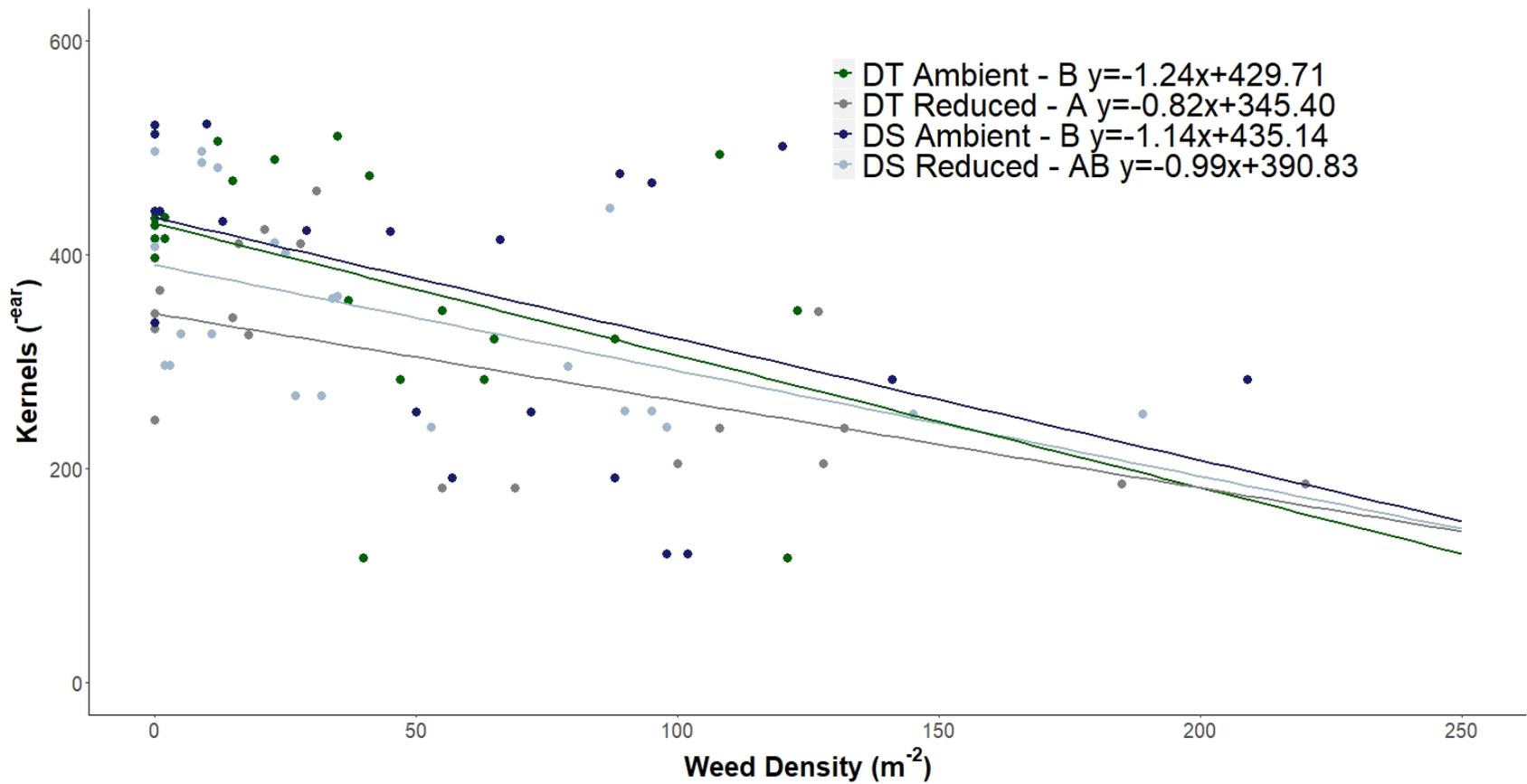


Figure 2.8. Corn kernels (<sup>-ear</sup>) as a function of weed density under two corn hybrids with differing levels of drought tolerance (DT=drought tolerant or DS=drought sensitive) and two precipitation levels (ambient or 70% reduced) in a two-year field experiment (2019-2020). Slopes obtained from regression lines labeled by the same capital letter are not statistically different ( $p \geq 0.05$ ).

Table 2.6. Mean (SE) corn height under three weed competition levels, two precipitation levels, and two corn hybrids with differing drought tolerance at four corn growth stages in a two-year field experiment (2019-2020).

Treatments <sup>1</sup>			Height			
			V4-V6*	V10-V12	Vt-R1	R3-R4
Hybrid	WP	Precipitation	cm			
DT	W1	F	12.54 (0.6) <i>aab</i> <sup>2,3</sup>	126.43 (1.0) <i>Aaa</i> <sup>2,3,4</sup>	170.13 (12.2) <i>Abaa</i> <sup>2,3,4</sup>	213.31 (12.0) <i>Aaba</i> <sup>2,3,4</sup>
DT	W1	R	13.85 (16.0) <i>bab</i>	125.57 (0.8) <i>Baa</i>	173.07 (12.0) <i>ACba</i>	213.26 (13.3) <i>Baca</i>
DT	W2	F	14.09 (1.0) <i>bc</i>	143.61 (0.7) <i>Abb</i>	179.37 (10.3) <i>ABbab</i>	221.27 (10.2) <i>Acac</i>
DT	W2	R	13.93 (14.9) <i>bc</i>	125.44 (10.1) <i>Bab</i>	173.61 (12.7) <i>ACabab</i>	210.03 (12.0) <i>Bbac</i>
DT	W3	F	13.14 (0.67) <i>aac</i>	124.17 (7.4) <i>Aca</i>	159.94 (13.3) <i>ABcc</i>	205.27 (12.4) <i>Adbc</i>
DT	W3	R	14.44 (16.2) <i>bac</i>	121.85 (7.8) <i>Bca</i>	158.17 (11.7) <i>ACdc</i>	200.15 (11.2) <i>Bdbc</i>
DS	W1	F	13.04 (1.0) <i>aac</i>	131.11 (14.4) <i>ABaa</i>	169.94 (20.4) <i>Bab</i>	216.95 (11.0) <i>Aabb</i>
DS	W1	R	14.03 (10.7) <i>bac</i>	131.94 (14.9) <i>ABab</i>	180.21 (7.5) <i>Cbb</i>	222.19 (7.4) <i>Cacb</i>
DS	W2	F	13.12 (0.8) <i>bac</i>	137.13 (15.1) <i>ABbb</i>	175.36 (6.5) <i>Bbb</i>	219.87 (6.2) <i>Acc</i>
DS	W2	R	13.81 (15.9) <i>bac</i>	128.51 (7.0) <i>ABab</i>	179.51 (9.7) <i>Cabb</i>	217.71 (11.0) <i>Cbb</i>
DS	W3	F	12.15 (0.8) <i>ab</i>	113.73 (10.1) <i>ABcc</i>	149.42 (6.1) <i>Bcd</i>	200.06 (6.0) <i>Add</i>
DS	W3	R	13.04 (13.1) <i>bb</i>	115.33 (7.4) <i>ABcc</i>	160.27 (9.8) <i>Cdd</i>	207.26 (9.0) <i>Cdd</i>

<sup>1</sup>Abbreviations: DT=drought tolerant; DS=drought sensitive; WP=weed competition; W1=weed free; W2=50% control; W3=no control; F=ambient; R=reduced.

<sup>2</sup>Means within the same column followed by the same lower-case letter are not statistically different ( $p \geq 0.05$ ) for a two-way interaction of weed competition and precipitation.

<sup>3</sup>Means within the same column followed by the same italicized lowercase letter are not statistically different ( $p \geq 0.05$ ) for a two-way interaction of weed competition and hybrid.

<sup>4</sup>Means within the same column followed by the same capital letter are not statistically different ( $p \geq 0.05$ ) for a two-way interaction of precipitation and hybrid.

\*Non-significant two-way interaction of hybrid and weed competition.

Table 2.7. Mean (SE) corn stem diameter under three weed competitions, two precipitation levels, and two corn hybrids with differing drought tolerance at four corn growth in a two-year field experiment (2019-2020).

Treatments <sup>1</sup>			Stem diameter			
			V4-V6	V10-V12	Vt-R1 <sup>+</sup>	R3-R4
Hybrid	WP	Precipitation	mm			
DT	W1	F	13.99 (0.38) <i>aa</i> <sup>2,3</sup>	25.56 (0.61) <i>aa</i> <sup>2,3</sup>	23.35 (0.31) <i>a</i> <sup>3</sup>	23.31 (0.53) <i>aa</i> <sup>2,3</sup>
DT	W1	R	13.94 (0.56) <i>aab</i>	23.89 (0.71) <i>ba</i>	23.34 (0.80) <i>ab</i>	21.83 (0.60) <i>ba</i>
DT	W2	F	15.51 (0.52) <i>bc</i>	26.45 (0.40) <i>aa</i>	25.79 (0.48) <i>ab</i>	23.47 (0.40) <i>aa</i>
DT	W2	R	14.34 (0.41) <i>abc</i>	23.18 (0.41) <i>bca</i>	22.84 (0.53) <i>ab</i>	21.56 (0.38) <i>ba</i>
DT	W3	F	14.19 (0.38) <i>a<sup>abc</sup></i>	23.68 (0.79) <i>cdb</i>	24.85 (2.68) <i>bc</i>	20.71 (0.98) <i>cb</i>
DT	W3	R	14.24 (0.55) <i>a<sup>abc</sup></i>	22.39 (0.58) <i>db</i>	21.17 (0.64) <i>bc</i>	19.66 (0.75) <i>cb</i>
DS	W1	F	14.09 (0.33) <i>aac</i>	25.34 (0.44) <i>aa</i>	25.69 (0.79) <i>a</i>	23.78 (0.62) <i>aa</i>
DS	W1	R	14.42 (0.79) <i>aac</i>	24.23 (0.65) <i>ba</i>	24.16 (0.74) <i>a</i>	21.70 (0.71) <i>ba</i>
DS	W2	F	14.51 (0.48) <i>bac</i>	25.86 (0.31) <i>aa</i>	25.77 (0.63) <i>a</i>	23.62 (0.52) <i>aa</i>
DS	W2	R	14.58 (0.73) <i>abac</i>	24.06 (0.60) <i>bca</i>	23.56 (0.87) <i>a</i>	21.86 (0.58) <i>ba</i>
DS	W3	F	13.60 (0.50) <i>ab</i>	22.54 (0.75) <i>cdb</i>	22.90 (1.71) <i>c</i>	19.71 (1.72) <i>cb</i>
DS	W3	R	13.62 (0.52) <i>ab</i>	21.80 (0.56) <i>db</i>	20.91 (0.57) <i>c</i>	19.63 (0.50) <i>cb</i>

<sup>1</sup>Abbreviations: DT=drought tolerant; DS=drought sensitive; WP=weed competition; W1=weed free; W2=50% control; W3=no control; F=ambient; R=reduced.

<sup>2</sup>Means within the same column followed by the same lowercase letter are not statistically different ( $p \geq 0.05$ ) for the two-way interaction of precipitation and weed competition.

<sup>3</sup>Means within the same column followed by the same italicized lowercase letter are not statistically different ( $p \geq 0.05$ ) for the two-way interaction of hybrid and weed competition.

<sup>+</sup>Non-significant two-way interaction of precipitation and weed competition.

Table 2.8. Mean (SE) corn growth stage under three weed competitions, two precipitation levels, and two corn growth stages with differing drought tolerance at five timings in a two-year field experiment (2019-2020).

Treatments <sup>1</sup>			June <sup>v</sup>	July (early) <sup>*+v</sup>	July (late) <sup>o</sup>	July (late) <sup>o*v</sup>	August <sup>v</sup>
Hybrid	WP	Precipitation	Corn growth stage				
DT	W1	F	V6 (0.13)aba <sup>2,3</sup>	V12 (0.92)Bb <sup>5,6</sup>	V15 (0.25)ABaa <sup>2,3,4</sup>	R1 (0.25)B <sup>6</sup>	R4 (0.19)aa <sup>2,3</sup>
DT	W1	R	V5 (0.13)aa	V12 (0.62)Bb	V15 (0.29)ABaba	R1 (0.00)B	R3 (0.35)ba
DT	W2	F	V5 (0.26)aa	V12 (0.67)Bb	V16 (0.29)ABbab	VT (0.25)B	R4 (0.26)ab
DT	W2	R	V5 (0.16)aa	V12 (0.78)Bb	V15 (0.41)ABaab	R1 (0.00)B	R3 (0.27)cb
DT	W3	F	V5 (0.13)ba	V11 (0.70)Ba	V13 (0.95)ABcc	VT (0.00)A	R3 (0.18)dc
DT	W3	R	V6 (0.20)aa	V11 (0.69)Ba	V14 (0.87)ABdc	VT (0.00)A	R3 (0.18)dc
DS	W1	F	V5 (0.00)aba	V12 (0.68)Ab	V16 (0.41)Aaab	R1 (0.25)B	R4 (0.26)aab
DS	W1	R	V6 (0.18)aa	V12 (0.78)Ab	V16 (0.48)Babab	R1 (0.25)B	R3 (0.33)bab
DS	W2	F	V6 (0.13)aa	V12 (0.63)Ab	V16 (0.29)ABab	R1 (0.00)B	R4 (0.19)aab
DS	W2	R	V6 (0.16)aa	V12 (0.78)Ab	V16 (0.58)Bab	R1 (0.25)B	R3 (0.30)cab
DS	W3	F	V5 (0.13)bb	V11 (0.88)Aa	V12 (1.08)Acd	VT (0.00)A	R3 (0.13)dc
DS	W3	R	V5 (0.19)ab	V11 (0.68)Aa	V14 (0.48)Bdd	VT (0.50)A	R3 (0.16)dc

Notes: July late timing was separated by years due to data collection not being the same between the two years.

<sup>1</sup>Abbreviations: DT=drought tolerant; DS=drought sensitive; WP=weed competition; W1=weed free; W2=50% control; W3=no control; F=ambient; R=reduced

<sup>2</sup>Means within the same column followed by the same lowercase letter are not statistically different ( $p \geq 0.05$ ) for the two-way interaction of weed competition and precipitation.

<sup>3</sup>Means within the same column followed by the same italicized lowercase letter are not statistically different ( $p \geq 0.05$ ) for the two-way interaction of weed competition and hybrid.

<sup>4</sup>Means within the same column followed by the same capital letter are not statistically different ( $p \geq 0.05$ ) for the two-way interaction of precipitation and hybrid.

Table 2.8. (cont'd),

<sup>5</sup>Means within the same column followed by the same bold lowercase letter are not statistically different ( $p \geq 0.05$ ) for the main effect of hybrid.

<sup>6</sup>Means within the same column followed by the same italicized capital letter are not statistically different ( $p \geq 0.05$ ) for the main effect of weed competition.

\*Non-significant two-way interaction of hybrid and weed competition ( $p \geq 0.05$ ).

∇Non-significant two-way interaction of hybrid and precipitation ( $p \geq 0.05$ ).

+Non-significant two-way interaction of precipitation and weed competition ( $p \geq 0.05$ ).

<sup>◇</sup>2019 growing season.

<sup>◊</sup>2020 growing season.

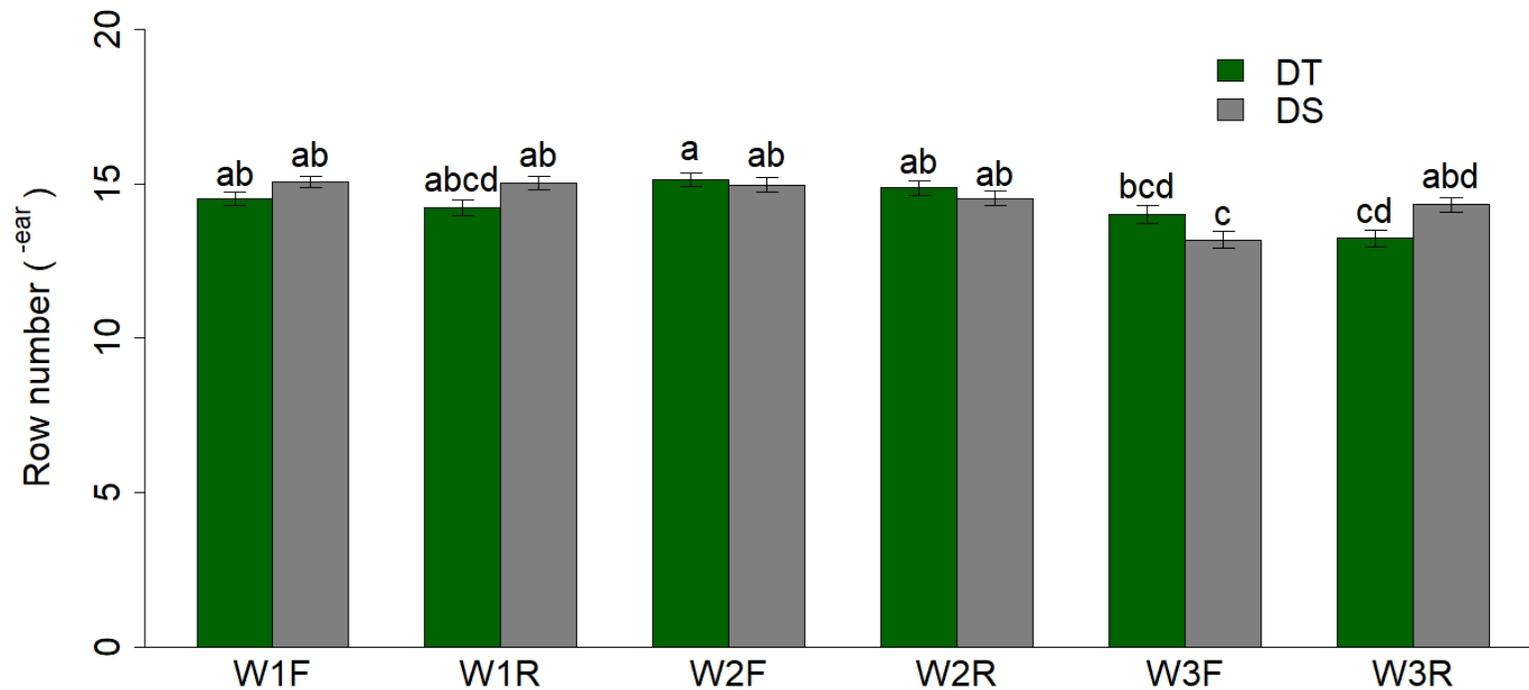


Figure 2.9. Mean (SE) row numbers (<sup>-ear</sup>) under three weed competitions, two precipitation levels, and two corn hybrids with differing drought tolerance in a two-year field experiment (2019-2020). Bars followed by the same lowercase letter are not statistically different for the three-way interaction of hybrid, weed, and precipitation ( $p \geq 0.05$ ).

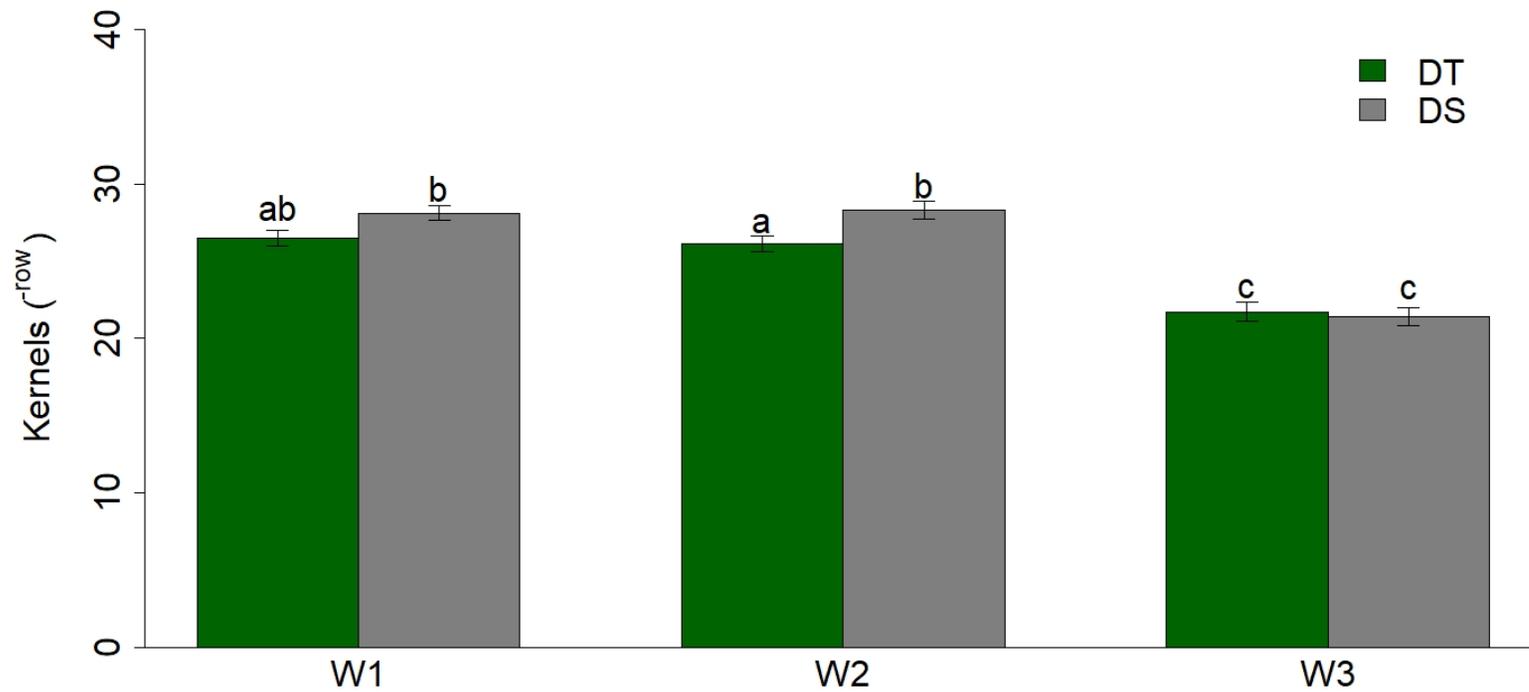


Figure 2.10. Mean (SE) kernels (<sup>-row</sup>) under three weed competitions and two hybrids in a two-year field experiment (2019-2020). Bars followed by the same lowercase letter are not statistically different for the two-way interaction of hybrid and weed competition ( $p \geq 0.05$ ).

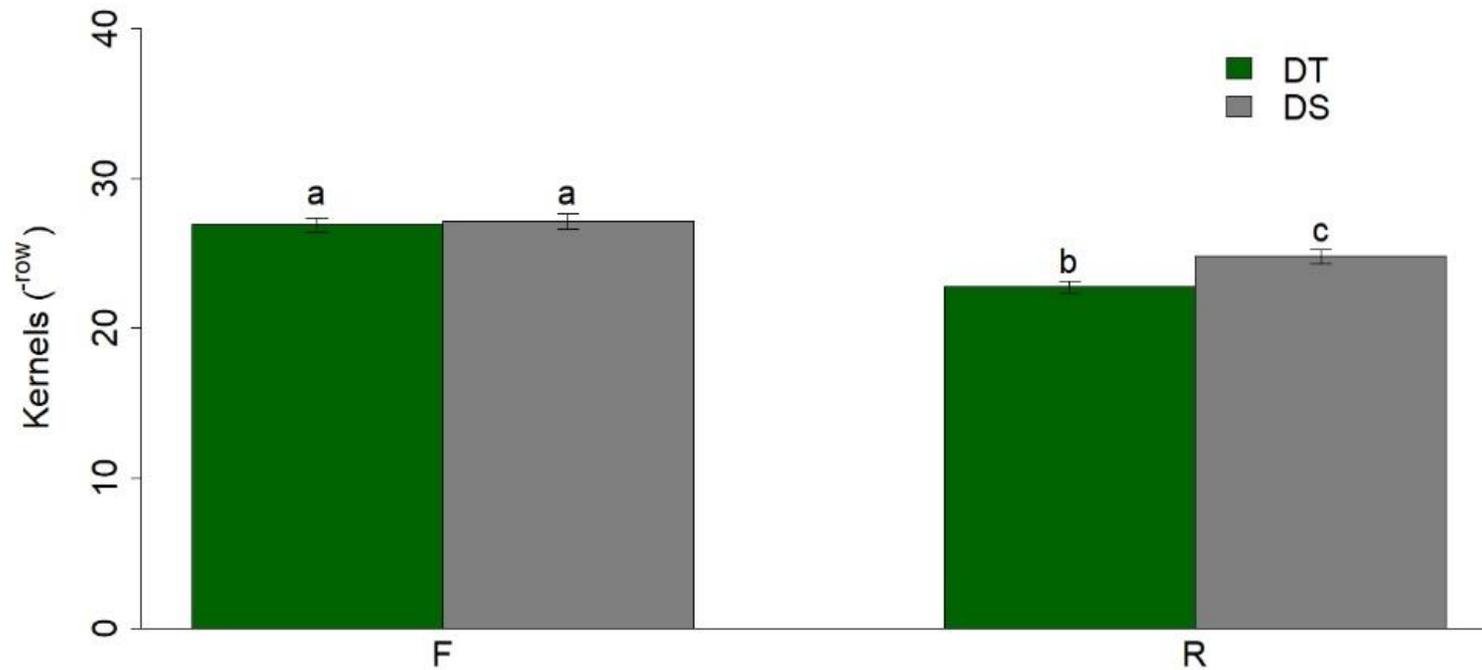


Figure 2.11. Mean (SE) kernels ( $^{-row}$ ) under two hybrids with differing drought and two precipitation levels in a two-year field experiment (2019-2020). Bars followed by the same lowercase letter are not statistically different for the two-way interaction of hybrid precipitation ( $p \geq 0.05$ ).

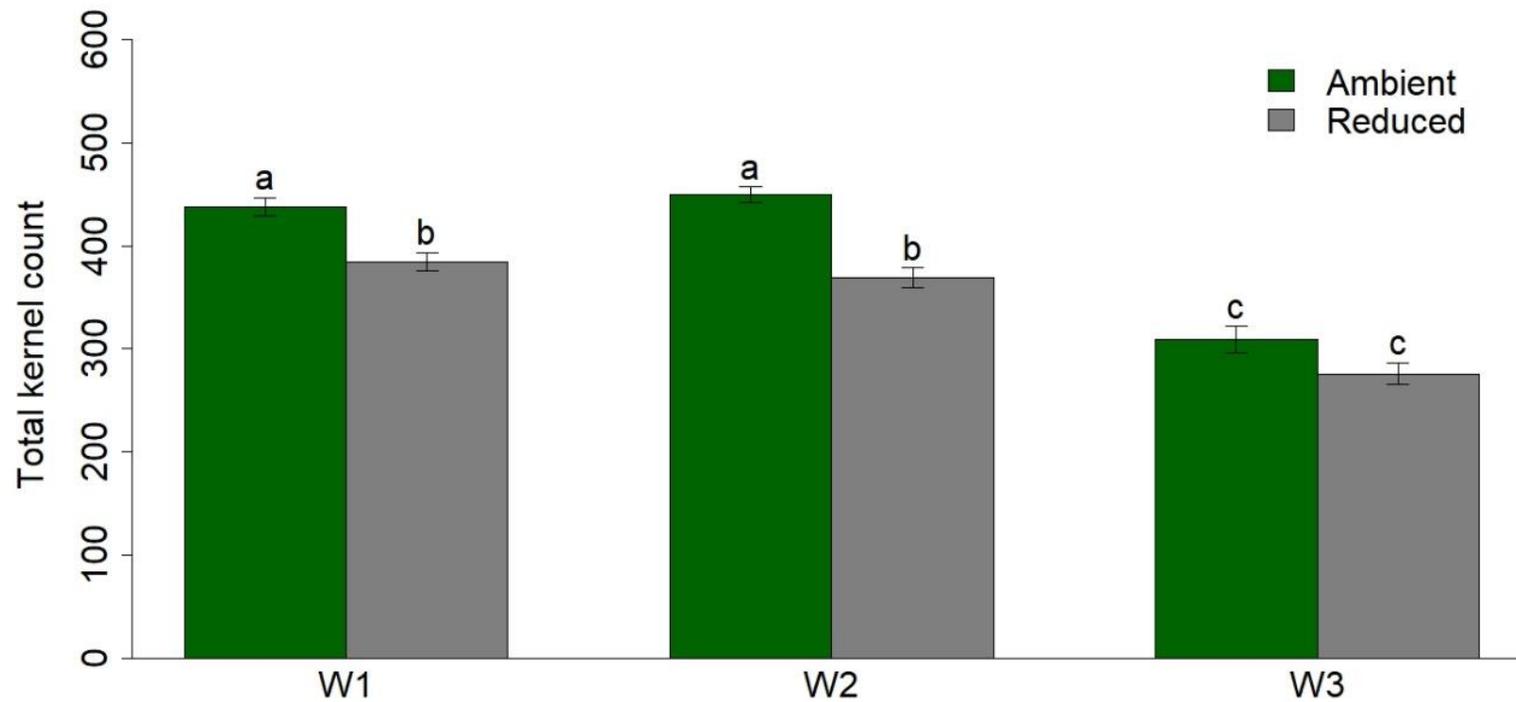


Figure 2.12. Mean (SE) total kernel count under three weed competitions and two precipitation levels in a two-year field experiment (2019-2020). Bars labeled by the same lowercase letter are not statistically different for the two-way interaction of hybrid and precipitation ( $p \geq 0.05$ ).

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## CHAPTER III

### IMPACT OF WATER STRESS AND VARYING PRESSURES ON DROUGHT TOLERANT CORN PERFORMANCE

#### Abstract

Water stress and weed competition are critical stressors during corn (*Zea mays* L.) development. Genetic improvements in corn have resulted in hybrids with greater tolerance to abiotic and biotic stressors; however, drought stress remains problematic. Therefore, with the expected change in precipitation throughout the Great Lakes Region, greenhouse experiments were conducted to evaluate water stress and weed competition on drought tolerant corn performance. The study followed a completely randomized design with four replications. Factorial combinations consisted of drought tolerant corn competition (presence or absence), water stress (100 or 50% volumetric water content (VWC)), and nine corn:common lambsquarters (*Chenopodium album* L.) densities. Corn and common lambsquarters growth parameters were measured 14 and 21 days after water stress initiation. To address the impact of reduced soil moisture and weed competition on corn and common lambsquarters growth parameters, photosynthetic response, and biomass; linear mixed effects and non-linear regression models were constructed in R. Common lambsquarters biomass was reduced by 46 and 50% under corn competition at two and four weeds  $\text{pot}^{-1}$  ( $p = 0.0003, 0.0004$ ). However, introducing crop competition under six and nine weeds  $\text{pot}^{-1}$  did not reduce common lambsquarters biomass ( $p = 0.90, 1.00$ ). Averaged across weed pressures, corn biomass was 22% less when grown under 50% compared to 100% VWC ( $p = 0.0003$ ). However, averaged across VWC, increasing weed competition from zero to two ( $p = 0.04$ ), four ( $p = <0.0001$ ), six ( $p = 0.0002$ ), or nine ( $p = 0.0002$ ) weeds  $\text{pot}^{-1}$  reduced biomass by 22, 38, 35, and 36%. Overall, water stress and common lambsquarters competition negatively affected the parameters measured in this study; however,

the magnitude of reduction is stronger under drought stress than increasing weed competition when water is not limiting. Therefore, field crop growers will need to modify current integrated weed management programs to maintain yield under future climate stress.

## Introduction

The projections change in climate for the Great Lakes Region will affect agricultural production in many ways. Since 1951 the Great Lakes Region has seen an increase in annual average air temperature of  $-16.5^{\circ}\text{C}$ , 14% increase in total precipitation, and a 35% increase in heavy precipitation events (GLISAP, 2017). Additionally, the Great Lakes Integrated Sciences and Assessments program projects yearly precipitation will increase; however, this precipitation will fall in heavy rainfall events, leaving more days per year that receive little or no precipitation.

Given these projections drought events may become more common in the Great Lakes Region. Water stress is a critical abiotic stress during corn development (Witt et al., 2012). Although corn is a  $\text{C}_4$  plant, resulting in its ability to photosynthesize when the stomates are closed (Lopes et al., 2011), drought conditions can significantly impact plant growth and phenology (Hatfield and Prueger, 2015). Water stress during the vegetative growth stages reduces stem and leaf cell expansion, resulting in shorter plants with less leaf area (Licht and Archontoulis, 2017). Additionally, during the reproductive growth stages, substantial yield loss can occur if water stress occurs during silking and grain fill (Claassen and Shaw, 1970).

In conjunction with water stress, high temperatures can significantly impact corn yield, specifically during the pollination phase (Hatfield and Prueger, 2015). Corn pollen viability decreased by 59% when exposed to temperatures above  $32^{\circ}\text{C}$  compared to  $27^{\circ}\text{C}$  (Herrero and Johnson, 1980). Further, extreme temperatures can impact the rate of maturity and senescence. Hatfield and Prueger (2015) reported when corn plants were grown under the extreme temperature of  $34^{\circ}\text{C}$  for 120 days, total vegetative biomass increased by 20% compared to total vegetative biomass of plants grown under  $30^{\circ}\text{C}$ ; however, yield decreased by 88%. Hatfield and Prueger (2015) also suggested that during the vegetative growth stages of corn, warmer daytime

temperatures do not negatively affect total vegetative biomass but do have strong negative consequences on grain yield.

Since the intensification of modern-day production agriculture began, drought tolerant corn hybrids have not been commercially available to protect against water deficiency stress caused by drought events. Recently, an emphasis towards genetic modification of crops, specifically corn, has led to an increase in research and development in many agriculture companies and universities (Bruns, 2019). DroughtGard™ corn was developed by Bayer Crop Science through the constitutive expression of cold shock protein B (cspB) from *Bacillus subtilis* to improve performance of corn under drought conditions (Wang et al., 2015). cspBs from the bacterial species *B. subtilis* bind to single-stranded nucleic acids, therefore acting as a RNA chaperone (Zeeb and Balbach, 2003). Functionally, cspB enables corn plants to decrease the rate of water absorption from the soil in dry conditions, therefore enabling the corn plant to withstand drought conditions for longer periods of time than conventional non-drought tolerant hybrids (Eisenstein, 2013).

Drought tolerant corn hybrids are often marketed as providing drought and heat tolerance in low-yielding environments where water is scarce (Newell et al., 2015). Currently there is limited research that has been conducted on drought tolerant corn hybrids. Studies conducted under limited irrigated environmental conditions in Kansas (Kisekka et al., 2015) and Mississippi (Bruns, 2019) reported no difference in water use efficiency and grain yield between the DroughtGard™ corn hybrid and a non-drought tolerant hybrid.

In addition to the abiotic stressors of moisture and temperature, a significant biotic stress is weed competition. Weeds are troublesome, aggressive, and competitive plants that share a similar trophic level with crops, allowing them to compete for scarce resources of water, light,

and nutrients (Ramesh et al., 2017). With the projected increase in erratic rainfall events, weed impacts on agriculture commodities are expected to change resulting in shifts of competition (Ramesh et al., 2017). Rodenburg et al. (2010) hypothesizes that during extended drought conditions and increased temperatures, C<sub>4</sub> weeds will outcompete C<sub>3</sub> weeds. Reasons for this shift surround photosynthetic efficiency, C<sub>4</sub> weeds have a higher water use efficiency than C<sub>3</sub>, thus C<sub>4</sub> weeds may become more dominant under dry environmental conditions (Singh et al., 2011).

Common lambsquarters (*Chenopodium album* L.) is a C<sub>3</sub> broadleaf summer annual weed that is common in many agricultural fields (Korres et al., 2016). Without crop competition, common lambsquarters is a prolific seed producer and can produce 75,600-150,400 seeds plant<sup>-1</sup>; however, in competition with corn, common lambsquarters seed production is reduced to 110-3,600 seeds plant<sup>-1</sup> (Colquhoun et al., 2001). Furthermore, in a list of the world's worst weeds by Holm et al. (1977), common lambsquarters was in the top 10 worst weeds. In competition with corn, common lambsquarters has been found to decrease yield by 12% with 4.9 plants per 1 m of row (Beckett et al., 1988).

In addition to crop plants, weeds are also subjected to drought conditions (Maganti et al., 2005). Common lambsquarters height was reduced by 55 and 29% under drought conditions (no water for 21 d after initial establishment period) in experimental runs 1 and 2 compared to continuously watered plants (Maganti et al., 2005). Furthermore, in the same study, common lambsquarters shoot dry weight was reduced by 28 and 64% under drought conditions compared to the non-drought treatment.

With future climate change projections, it is important that producers have an action plan to help mitigate potential negative impacts on crop growth, weed competition, and yield. With

the introduction of drought tolerant corn hybrids into the market, it is still unclear how these new hybrids will perform under drought conditions and weed pressure in the Great Lakes Region. Therefore, the objective of this research was to evaluate drought tolerant corn hybrid and common lambsquarters growth and physiology under two watering regimes and nine competition scenarios.

### **Materials and Methods**

Greenhouse experiments were conducted February-April 2020 at Michigan State University in East Lansing, MI. The study followed a completely randomized design with four replications and two experimental runs were performed. Treatments included factorial combinations of two levels of water stress, 100 and 50% soil volumetric water content (VWC), and nine corn:common lambsquarters densities, 1:0, 1:2, 1:4, 1:6, 1:9, 0:2, 0:4, 0:6, and 0:9.

Common lambsquarters seed was collected from the Michigan State University Beef Farm in 2018, threshed, and stored at 5°C until experiments began. To break dormancy, seeds were treated with a cold-water bath for 7 d prior to the experiment. The corn hybrid used in this study was DKC47-27 Droughtgard Double Pro® (Bayer Crop Science, Whippany, NJ U.S.). Soil used for the study was a 50:50 mixture of greenhouse media (SUREMIX, Michigan Grower Products, INC™ Galesburg, MI) and sterilized field soil. Field soil was screened with a 6 mm sieve for uniform consistency. The final soil mixture was a sandy loam with a pH of 7.4, 15.4% organic matter, 64.9% sand, 9.6% silt, and 10.1% clay.

Soil water holding capacity was determined following the methods of Sarangi et al. (2016). Final water holding capacity was calculated using the following equation (equation 1):

$$WC = [W_w - W_d]/d \quad [1]$$

where  $WC$  is the total water content,  $W_w$  is the weight of the saturated pot of soil,  $W_d$  is the weight of the dry pot of soil, and  $d$  is the density of water (Sarangi et al., 2016). Greenhouse temperature was set to 27°C (diurnal range 25-29°C) with a 16 hr photoperiod.

To ensure both common lambsquarters and corn plants emerged at the same time, common lambsquarters seeds were planted four days before corn into 20 cm wide and 14 cm deep (2.8 L) pots. Common lambsquarters seedlings were thinned to desired densities once plants reached the cotyledon stage. To ensure uniform germination, pots were watered to 100% soil VWC until corn reached the two-leaf stage and common lambsquarters reached 2 cm. Volumetric water content was monitored daily using a Field Scout TDR 300 Meter and then watered accordingly to achieve desired moisture levels. In both experimental runs corn height and growth stage and weed height and leaf number were measured every 7 d. Corn photosynthetic efficiency was measured every 14 d in both experimental runs. Above ground biomass of corn and weeds were harvested at 3 wk (run 1) and 4 wk (run 2) after water stress began, dried at 60°C, and weighed.

### **Statistical Analyses**

Data from experimental runs were combined after examining side-by-side box plots of the residuals, as well as a Levene's test for unequal variances. Normality assumptions were assessed by examining normal probability plots of the residuals and histograms. Growth parameters of corn and common lambsquarters were analyzed using non-linear regression in the drc package in R (2021) following the methods outlined in Knezevic et al. (2007). Three parameter log-logistic models were fit to weed height and leaf number (equation 2) and three parameter Weibull models were fit to corn growth stage and height (equation 3) using the drc modelFit function (Table 3.1).

$$Y = \frac{d}{1 + \exp[b(\log x - e)]} \quad [2]$$

$$Y = d \exp \{- \exp[b(\log x - e)]\} \quad [3]$$

For all equations,  $Y$  is the response variable (height, leaf number or growth stage),  $x$  is the number of days after water stress treatment initiation,  $c$  and  $d$  are the lower and upper limits, respectively,  $b$  is the relative slope around  $e$ , and  $e$  is the inflection point (Streibig, 1988).

Additionally, to address the impact of reduced soil moisture and weed competition on corn photosynthetic response, corn biomass, and weed biomass linear mixed effects models were constructed using the lmer function from the lme4 package in R (2021). Weed pressure, corn competition, and soil VWC were considered fixed effects and replication and run were considered random effects. Differences in means were further investigated using Tukey's HSD post hoc test in the emmeans package in R (2021).

## Results and Discussion

### Common lambsquarters

#### *Height*

Corn competition, increasing weed pressure, and water stress did not decrease the days needed to reduce height by 10 or 20% (Table 3.2). Interestingly, with six weeds pot<sup>-1</sup>, without corn competition, under 50% VWC, common lambsquarters plants reached a 40% reduction in height three days faster than under 100% VWC (Table 3.2,  $p = 0.07$ ). However, overall, holding common lambsquarters density constant, without corn competition, decreasing VWC did not decrease the days needed to reduce height by 40% (Table 3.2). Furthermore, holding common lambsquarters density constant, with corn competition, decreasing VWC did not affect the number of days needed to reduce common lambsquarters height by 40% (Table 3.2). Without corn competition, common lambsquarters plants grown under 50% VWC and 4 plants pot<sup>-1</sup>

reached a 40% reduction in height 4.2 days faster than common lambsquarters plants grown under 100% VWC and 6 plants pot<sup>-1</sup> (Table 3.2, p = 0.0003). Furthermore, common lambsquarters plants grown under 9 plants pot<sup>-1</sup>, no corn competition, and 50% VWC reached a 40% reduction in height 2.9 days faster than common lambsquarters plants grown under the same density, but with the addition of corn competition and 100% VWC (Table 3.2, p = 0.02).

Overall, these results demonstrate that reducing the available water for common lambsquarters growth is a larger stress than increasing inter- or intraspecific competition under ample water conditions. To our knowledge, there has not been a study conducted evaluating water stress and drought tolerant crop competition on weed height. However, Maganti et al. (2005) reported that common lambsquarters height was reduced 29 to 55% with no crop competition under drought conditions. Additionally, Sarangi et al. (2016) reported that decreasing VWC by 50% decreased common waterhemp (*Amaranthus rudis* J. D. Sauer) height by 10% compared to no water stress without crop competition.

#### *Leaf Number*

Corn competition, increasing weed pressure, and water stress did not decrease the days needed to reach a 10% reduction in leaf number (Table 3.2). Holding common lambsquarters density constant, with or without corn competition, reducing VWC by 50%, had no impact on the days needed to reduce leaf number by 20% (Table 3.2). However, without corn competition, common lambsquarters plants grown under four weeds pot<sup>-1</sup> and 50% VWC reached a 20% reduction in leaf number 2.6 and 2.2 days faster than common lambsquarters grown under six or nine weeds pot<sup>-1</sup> and 100% VWC (Table 3.2, p = 0.02 and 0.07). Interestingly, common lambsquarters plants grown under four weeds pot<sup>-1</sup>, corn competition, and 50% VWC, reached a 20% reduction in total leaf production 2.9 days faster than common lambsquarters plants grown

under nine weeds  $\text{pot}^{-1}$ , corn competition, and 100% (Table 3.2,  $p = 0.07$ ). Additionally, common lambsquarters plants grown under four plants  $\text{pot}^{-1}$ , no corn competition, and 50% VWC reached a 20% reduction in height 2.9 days faster than common lambsquarters plants grown under the same density, but under corn competition and 100% VWC (Table 3.2,  $p = 0.02$ ). Furthermore, common lambsquarters plants grown under nine plants  $\text{pot}^{-1}$ , no corn competition, and 50% VWC reached a 20% reduction in height 2.9 days faster than common lambsquarters plants grown under the same density, but under corn competition and 100% VWC (Table 3.2,  $p = 0.09$ ).

Holding weed pressure constant, with no corn competition, and decreasing VWC by 50% did not affect the time needed to reach a 40% reduction in common lambsquarters leaf number (Table 3.2). However, common lambsquarters plants grown with four weeds  $\text{pot}^{-1}$ , crop competition, and 50% VWC reached a 40% reduction in leaf number 4 days faster than common lambsquarters plants grown under the same crop and weed density, but under 100% VWC (Table 3.2,  $p = 0.03$ ). Additionally, holding weed pressure constant at nine weeds  $\text{pot}^{-1}$ , increasing VWC by 50% and adding corn competition increased the time required to reach a 40% reduction by 5 days (Table 3.2,  $p = 0.02$ ). A similar trend occurred with four weeds  $\text{pot}^{-1}$  resulting in an increase in the time required to reach a 40% reduction in leaf number by 7 days (Table 3.2,  $p = <0.0001$ ). Finally, without crop competition, common lambsquarters plants grown under four weeds  $\text{pot}^{-1}$  and 50% VWC reached a 40% reduction in leaf number 4.4 and 5.8 days faster than common lambsquarters grown under six and nine weeds  $\text{pot}^{-1}$  and 100% VWC (Table 3.2,  $p = <0.0001$  and  $<0.0001$ ).

Overall, these results demonstrate that introducing water stress decreases the rate of common lambsquarters leaf production. To our knowledge, there has not been a study conducted evaluating water stress and drought tolerant crop competition on common lambsquarters leaf

production. Additionally, our results highlight that increasing weed or crop competition had little impact on overall leaf production compared to water stress. Sarangi et al. (2016) reported that decreasing soil moisture by 50% decreased common waterhemp leaf production by 30% compared to no water stress without crop competition.

### *Biomass*

Reducing VWC by 50% did not reduce common lambsquarters biomass averaged across levels of corn and weed competition ( $p = 0.18$ ). However, there was a significant two-way interaction between corn competition and weed pressure ( $p = 0.05$ ). Without corn competition, increasing weed pressure from two to four weeds  $\text{pot}^{-1}$  did not reduce weed biomass ( $p = 0.98$ ). However, without corn competition, increasing weed pressure from two weeds  $\text{pot}^{-1}$  to six and nine weeds  $\text{pot}^{-1}$ , decreased weed biomass by 44 and 56% (Figure 3.1,  $p = 0.0005$  and  $<0.0001$ ). Furthermore, without corn competition, increasing weed pressure from four weeds  $\text{pot}^{-1}$  to six and nine weeds  $\text{pot}^{-1}$ , decreased weed biomass by 38 and 51% (Figure 3.1,  $p = 0.01$  and  $0.0002$ ). Increasing weed pressure from six to nine weeds  $\text{pot}^{-1}$  did not negatively impact weed biomass (Figure 3.1,  $p = 0.95$ ).

Under corn competition, increasing weed pressure did not reduce weed biomass (Figure 3.1,  $p \geq 0.05$ ). Additionally, common lambsquarters biomass was reduced by 46% and 50% when corn competition was introduced under two and four weeds  $\text{pot}^{-1}$ , respectively (Figure 3.1,  $p = 0.0003$  and  $0.0004$ ). However, introducing crop competition under six and nine weeds  $\text{pot}^{-1}$  did not reduce common lambsquarters biomass (Figure 3.1,  $p = 0.90$  and  $1.00$ ). Overall, these results demonstrate that increasing corn competition does not reduce weed biomass under high weed pressures but does decrease weed biomass under lower weed pressures. These findings support the results previously discussed regarding common lambsquarters height and leaf

number. Similar results were reported by Sarangi et al. (2016) in which reducing VWC by 75%, common waterhemp plant height, leaves per plant, and aboveground biomass decreased without corn competition. However, Chahal et al. (2018) reported that reduced soil moisture did not impact Palmer amaranth (*Amaranthus palmeri* S. Watson) leaf production, but did reduce aboveground biomass by 35% compared to the control of no water stress.

Furthermore, we hypothesize that if allowed to grow until maturity, similar results as Sarangi et al. (2016) would be reported. Therefore, future research should be conducted for a longer duration using larger pots to encompass common lambsquarters complete life cycle to confirm our study's findings. Overall, it is evident that water stress plays a larger role in common lambsquarters physiological development than weed or crop competition during the first 21 days of growth.

## Corn

### *Height*

Increasing weed pressure and reducing VWC by 50% did not decrease the number of days required to reduce corn height by 10% (Table 3.3). However, corn in competition with zero ( $p = 0.09$ ), two ( $p = 0.007$ ), and nine ( $p = 0.07$ ) weeds  $\text{pot}^{-1}$  under 100% VWC reached a 20% reduction in height 1.2, 2.1, and 2.8 days faster than under 50% VWC, respectively (Table 3.3). Additionally, corn grown without weed competition under 100% VWC reached a 20% reduction in height 1.8, 2.0, and 3.7 days faster than corn grown under 50% VWC and two ( $p = 0.03$ ), four ( $p = 0.05$ ), or six ( $p = 0.004$ ) weeds  $\text{pot}^{-1}$  (Table 3.3). Furthermore, corn in competition with six weeds  $\text{pot}^{-1}$  under 50% VWC reached a 20% reduction in height 2.6 and 2.9 days later than corn in competition with four ( $p = 0.03$ ) and nine ( $p = 0.009$ ) weeds  $\text{pot}^{-1}$  under 100% VWC (Table 3.3).

Holding weed competition constant at zero ( $p = 0.68$ ), two ( $p = 0.13$ ), and four ( $p = 0.46$ ) weeds  $\text{pot}^{-1}$  decreasing VWC by 50% did not modify the number of days required to reach a 40% reduction in height (Table 3.3). However, holding weed competition constant at six ( $p = 0.03$ ) and nine ( $p = 0.07$ ) weeds  $\text{pot}^{-1}$  decreasing VWC by 50%, increased the number of days required to reach a 40% reduction in height by 17.2 and 21 days, respectively (Table 3.3). Additionally, decreasing VWC by 50% and increasing weed pressure from two to six weeds  $\text{pot}^{-1}$  ( $p = 0.01$ ) and four to nine weeds  $\text{pots}^{-1}$  ( $p = 0.02$ ), increased the number of days required to reach a 40% reduction in height by 18 and 26 days, respectively (Table 3.3).

Drought impacts on corn growth are well documented (Cakir, 2004; Ge et al., 2012; Licht and Archontoulis, 2017). From our results, drought tolerant corn reaches a 20 and 40% reduction in height faster under 100% than 50% VWC which suggests that under a 50% VWC, drought tolerant corn buffers weed competition better than under 100% VWC. To our knowledge, there has not been a study conducted evaluating water stress and weed competition on drought tolerant corn height. However, Licht and Archontoulis (2017) reported that water stress during the vegetative growth stages reduces stem and leaf cell expansion, resulting in shorter plants. Additionally, Cakir (2004) reported that water stress during the early vegetative growth stages of corn decreases corn height. Furthermore, research by Ge et al. (2012) reported that in the field among three increasing levels of water stress treatments (33, 55, and 75% VWC), differences in plant height were not detected 21 days after planting. In contrast, our results demonstrate that corn height is reduced within two weeks of water stress initiation (Table 3.3).

#### *Growth Stage*

Increasing weed pressure and water stress did not impact the number of days needed to reduce corn growth stage by 10, 20, or 40% (Table 3.3). Additionally, averaged across weed

pressures and VWC's, a 10, 20, and 40% reduction in corn growth stage occurred at approximately two, three, and six days after water stress initiation (Table 3.3). Our results are supported by those reported in Hatfield and Prueger (2015). Specifically increasing temperature by 4°C during the pollination stage did not affect leaf collar development among three water treatments (50, 100, and 125%) (Hatfield and Prueger, 2015). Although Hatfield and Prueger (2015) reported that corn leaf collars were not impacted, grain-fill was reduced, which can cause implications on final crop yield.

### *Biomass*

Averaged across weed pressures, corn biomass was reduced by 22% when grown under 50% VWC compared to 100% VWC (Figure 3.2,  $p = 0.0003$ ). Additionally, averaged across VWC, there was no difference in corn biomass when grown with two, four, six, and nine weeds pot<sup>-1</sup> (Figure 3.2). However, averaged across VWC, increasing weed competition from zero to two ( $p = 0.04$ ), four ( $p = <0.0001$ ), six ( $p = 0.0002$ ), or nine ( $p = 0.0002$ ) weeds pot<sup>-1</sup> biomass was reduced by 22, 38, 35, and 36% (Figure 3.2).

Overall, these results indicate that reduced soil moisture and high weed densities decreases corn biomass. However, the magnitude of corn biomass reduction is stronger under drought stress than increasing levels of weed competition when water is not limiting. Ge et al. (2012) reported corn biomass was reduced by 68% when grown under water stress (55% reduction in soil moisture). The reduction we observed in this study was not as severe, potential reasons include the length of the study (21 vs. 70 days) and field vs. greenhouse conditions. Specifically, our study evaluated impacts of drought and weed competition on vegetative growth, it is well known that drought during the reproduction growth stage of silking can cause yield decreases of 55% (Claassen and Shaw, 1970). Additionally, crop-weed interactions will vary

depending on other altered climatic conditions not evaluated in this study including temperature and soil type (Singh et al., 2011). Photosynthetic pathways also play a role in weed competitiveness under water stress. For example, C<sub>4</sub> species such as kochia (*Bassia scoparia* (L.) A. J. Scott) and Russian thistle (*Salsola tragus* L.), have been found to be highly competitive under reduced soil moistures (Wiese and Vandiver, 1970). In contrast, common lambsquarters utilized in this study is a C<sub>3</sub> broadleaf summer annual weed (Korres et al., 2016).

### *Photosynthesis*

Increasing weed pressure ( $p = 0.58$ ) and water stress ( $p = 0.38$ ) nor their interaction ( $p = 0.68$ ) negatively impacted Phi2 levels 14 days after treatment (Table 3.4). However, there was a significant main effect of weed pressure ( $p = 0.03$ ) and VWC ( $p = 0.05$ ) on Phi2 levels 21 days after treatment. Furthermore, at 21 days, decreasing VWC by 50% reduced corn Phi2 levels by 6.6% compared to 100% VWC averaged across weed densities (Table 3.4,  $p = 0.05$ ).

Additionally, at 21 days, increasing weed pressure from zero weeds  $\text{pot}^{-1}$  to four ( $p = 0.02$ ) or six ( $p = 0.05$ ) weeds  $\text{pot}^{-1}$  increased Phi2 levels by 16 and 15% respectively, averaged across VWC (Table 3.4). Phi2, or quantum yield of photosystem II photochemistry, is the percentage of incoming light that goes into photosystem II, which is a measure of photosynthetic efficiency (Kramer et al., 2004). Under water limiting conditions, results from this study demonstrate that drought tolerant corn photosynthetic efficiency is reduced. Previous research has also concluded that drought stress significantly reduced Phi2 levels in cowpea (*Vigna unguiculata* L. Walp.) (Mwale et al., 2017). However, increasing weed competition increased photosynthetic efficiency, therefore suggesting that drought tolerant corn hybrids will compete well with increasing weed densities.

Increasing weed pressure ( $p = 0.69$ ) and water stress ( $p = 0.26$ ) nor their interaction ( $p = 0.71$ ) negatively impacted PhiNPQ levels 14 days after treatment (Table 3.4). However, there was a significant main effect of weed pressure ( $p = 0.05$ ) and VWC ( $p = 0.02$ ) on PhiNPQ levels 21 days after treatment. Furthermore, at 21 days, decreasing VWC by 50% increased corn PhiNPQ by 20% compared to 100% VWC averaged across weed densities (Table 3.4,  $p = 0.02$ ). However, at 21 days, increasing weed competition from zero weeds  $\text{pot}^{-1}$  to four ( $p = 0.05$ ) or six ( $p = 0.09$ ) weeds  $\text{pot}^{-1}$  decreased PhiNPQ levels by 28 and 25%, respectively averaged across VWC (Table 3.4). PhiNPQ, or quantum yield of non-photochemical energy, measures energy loss via down regulation of photochemistry (Kramer et al., 2004). These results are in accordance with Phi2 in which drought reduced photosystem II efficiency thus leading to an increase in energy loss. Furthermore, the reduction in energy loss under increasing weed density is supported by the increase in photosystem II efficiency discussed above.

Increasing weed pressure ( $p = 0.99$ ) and reducing VWC by 50% ( $p = 0.21$ ) nor their interaction ( $p = 0.59$ ) negatively affected PhiNO levels 14 days after treatment (Table 3.4). However, 21 days after treatment, the main effect of VWC was significant ( $p = 0.008$ ), but the main effect of weed pressure ( $p = 0.30$ ) or their interaction ( $p = 0.47$ ) were not significant. Specifically, reducing VWC by 50% ( $p = 0.008$ ) decreased PhiNO levels by 9%, averaged across weed densities (Table 3.4). PhiNO is the quantum yield of other unregulated processes (Kramer et al., 2004). Unlike Phi2 and PhiNPQ increasing weed competition had no impact on PhiNO levels 21 days after treatment.

Overall the PhotosynQ instrument allows for the evaluation of sensitive indicators of various photosynthetic parameters and the ability to view the onset of photoinhibition and photodamage that may be caused by plant stress (Baker and Rosenqvist, 2004). To our

knowledge, there has not been a study conducted evaluating photosynthetic parameters on drought stressed drought tolerant corn hybrids and increasing weed competition using the PhotosynQ instrument. However, previous research has been conducted on the photosynthetic response of drought stressed drought tolerant corn hybrids using a Li-COR meter. A greenhouse study conducted by Wijewardana et al. (2017) reported that photosynthetic rates measured five times during the study declined in drought tolerant corn hybrids as soil moisture levels decreased. However, our results demonstrate that for the first 14 days of growth, reduced soil moisture and weed competition do not modify corn photosynthesis. However, after 21 days, reduced soil moisture decreased Phi2 and PhiNO levels, while increasing PhiNPQ. In contrast, increasing weed pressures increased phi2 while decreasing PhiNPQ and had no impact on PhiNO levels. It would be interesting to evaluate if our results of drought and weed competition modifying photosynthetic efficiency of vegetative drought tolerant corn impacts photosynthesis at later growth stages to fully understand how reduced soil moisture and weed competition impacts drought tolerant corn photosynthesis.

The projected changes in climate for the Great Lakes Region will affect agricultural production in many ways. Since 1951, the Great Lakes Region has seen an increase in annual average air temperature of  $-16.5^{\circ}\text{C}$ , 14% increase in total precipitation, and a 35% increase in heavy precipitation events (GLISAP, 2017). Given these projections, drought events may become more common in the Great Lakes Region. From these results, we can conclude that soil moisture stress plays a larger role in reducing common lambsquarters height and leaf number than drought tolerant corn competition. Additionally, drought tolerant corn competition had no impact on common lambsquarters biomass at high weed densities but did decrease common lambsquarters biomass at low weed densities. We attribute this difference to the fact that at the

higher weed densities, there is already so much stress occurring, that the additional stress of corn competition has little additional negative impact on biomass. However, corn reached a 40% reduction in height faster under 100% VWC than 50% which suggests that under a 50% VWC, drought tolerant corn buffers weed competition better than under 100% VWC. This effect is opposite in common lambsquarters in which it reaches reductions faster under reduced soil moisture which suggests that common lambsquarters responds to reduced soil moisture more than drought tolerant corn. Furthermore, reduced soil moisture decreased corn biomass 21 days after stress initiation. Additionally, any level of weed competition decreased corn biomass. Interestingly, reduced soil moisture decreased corn Phi2 levels, but increases PhiNPQ levels, thus leading to net energy loss. However, due to this study only being conducted for 21 days, future research should be conducted until natural plant senescence to fully understand how reduced soil moisture and weed competition impact growth and photosynthetic parameters of drought tolerant corn hybrid. Additionally, future research should include multiple levels of water stress outside those measured in this study, as well as evaluate different durations of water stress.

This study was the first step in identifying how climate will influence weed biology and drought tolerant corn production. Although genetic improvements to corn have been made resulting in corn with increased levels of drought tolerance it is evident weed competition under reduced soil moisture conditions will detrimentally impact yield. Therefore, taking proactive integrated weed management steps by identifying the corn-weed competition principles investigated in this study will allow field crop growers across the Great Lakes Region to integrate weed biology and crop physiology in the development of integrated economically viable weed management programs under future climate stress.

## **APPENDIX**

Table 3.1. List of models used for greenhouse growth parameters. Models were chose using the modelFit function in R (2021).

Growth parameter	Model	Model fit
Relative corn height	W2.3	p=0.18
Relative corn growth stage	W2.3	p=0.10
Relative weed height	LL.3	p=0.20
Relative weed leaf number	LL.3	p=0.96

Table 3.2. Mean (SE) days required to reduce common lambsquarters height and leaf number by 10, 20, and 40%, under two drought tolerant corn densities, four weed pressures, and two soil volumetric water content (VWC) levels in a greenhouse study. Percent reductions and probability values were calculated by ED and EDcomp functions, respectively, in the drc package in R (2021).

Treatments			Relative height <sup>1</sup>			Relative leaf number <sup>1</sup>		
Corn	Weed	VWC	10%	20%	40%	10%	20%	40%
Plants pot <sup>-1</sup>		%	Days			Days		
0	2	100	11.2 (4.70)a <sup>2</sup>	12.2 (3.28)a <sup>2</sup>	13.5 (1.19)abcd <sup>2</sup>	11.5 (4.59)a <sup>2</sup>	12.4 (3.14)abcd <sup>2</sup>	13.5 (1.07)abcd <sup>2</sup>
0	2	50	6.9 (1.50)a	9.0 (1.39)a	12.3 (1.09)abcde	7.3 (1.15)a	9.5 (1.04)abc	13.0 (0.80)ab
0	4	100	5.8 (0.96)a	7.4 (0.93)a	10.1 (0.84)ef	6.9 (0.94)a	8.6 (0.87)ab	11.4 (0.71)ae
0	4	50	5.9 (0.88)a	7.4 (0.86)a	9.6 (0.80)f	6.1 (0.84)a	8.0 (0.80)a	10.9 (0.68)e
0	6	100	7.3 (1.75)a	9.7 (1.59)a	13.8 (1.17)ac	7.8 (1.28)a	10.6 (1.13)bcd	15.3 (0.84)cdfg
0	6	50	6.1 (1.11)a	7.9 (1.06)a	10.8 (0.91)bef	6.7 (1.07)a	9.1 (0.99)abc	13.3 (0.80)bc
0	9	100	5.8 (1.32)a	8.3 (1.27)a	12.5 (1.07)abcd	6.8 (1.40)a	10.2 (1.27)bcd	16.7 (1.12)fghi
0	9	50	5.6 (1.08)a	7.6 (1.05)a	10.9 (0.92)bef	5.8 (1.36)a	9.3 (1.29)abc	16.2 (1.26)fghi
1	2	100	7.9 (1.82)a	9.8 (1.62)a	12.7 (1.16)abcd	7.3 (1.15)a	10.0 (1.05)bcd	14.6 (0.82)bcdf
1	2	50	6.4 (1.56)a	8.9 (1.48)a	13.2 (1.18)acd	6.7 (1.21)a	9.5 (1.12)abcd	14.5 (0.92)bcdf
1	4	100	5.3 (1.49)a	8.2 (1.47)a	13.8 (1.31)acd	7.1 (1.54)a	10.9 (1.37)bcd	18.3 (1.40)hij
1	4	50	5.6 (1.09)a	7.6 (1.06)a	11.0 (0.93)bdef	6.5 (1.15)a	9.3 (1.08)abc	14.3 (0.88)bcdf
1	6	100	6.5 (1.41)a	8.8 (1.33)a	12.5 (1.06)abcd	6.9 (1.43)a	10.4 (1.29)bcd	17.1 (1.17)ghi
1	6	50	5.8 (1.15)a	7.8 (1.11)a	11.2 (0.95)abdef	6.6 (1.31)a	9.7 (1.21)abcd	15.4 (0.98)dfgh
1	9	100	6.7 (1.57)a	9.3 (1.46)a	13.8 (1.12)c	7.7 (1.87)a	12.2 (1.56)d	21.3 (2.25)j
1	9	50	5.3 (1.33)a	7.8 (1.31)a	12.5 (1.13)abcd	7.4 (1.78)a	11.4 (1.54)cd	19.1 (1.57)ij

<sup>1</sup>Percent reduction data are days relative to the control of no corn competition, two weeds, and 100% VWC.

<sup>2</sup>Means within the same column followed by the same lowercase letter are not statistically different ( $p \geq 0.1$ ).

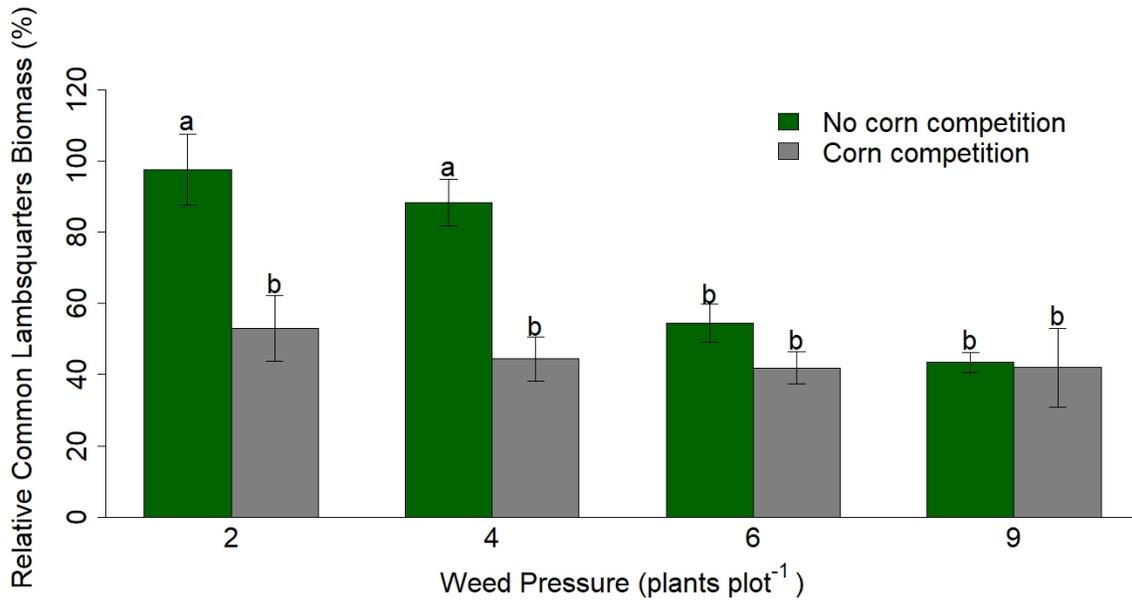


Figure 3.1. Mean (SE) relative common lambsquarters biomass, impacted by drought tolerant corn competition, four weed pressures, and averaged across two (50 and 100%) soil volumetric water contents (VWC) in a greenhouse study. Biomass reduction data are relative to the control of no corn competition, two weeds, and 100% VWC. Bars labeled by the same lowercase letter are not statistically different ( $p \geq 0.05$ ).

Table 3.3. Mean (SE) days required to reduce drought tolerant corn height and growth stage by 10, 20, and 40%, under four weed pressures and two soil volumetric water content (VWC) levels in a greenhouse study. Percent reductions and probability values were calculated by ED and EDcomp functions, respectively, in the drc package in R (2021).

Treatments			Relative height <sup>1</sup>			Relative growth stage <sup>1</sup>		
Corn	Weed	VWC	10%	20%	40%	10%	20%	40%
Plants pot <sup>-1</sup>		%	Days					
1	0	100	5.7 (2.55)a <sup>2</sup>	14.4 (6.78)abc <sup>2</sup>	40.1 (20.54)a <sup>2</sup>	1.9 (0.35)a <sup>2</sup>	3.1 (0.42)a <sup>2</sup>	5.5 (0.42)a <sup>2</sup>
1	0	50	6.3 (2.74)a	15.6 (7.13)def	42.3 (21.13)ab	2.2 (0.34)a	3.7 (0.40)a	6.3 (0.41)a
1	2	100	5.6 (2.48)a	14.1 (6.67)a	39.5 (20.42)a	2.2 (0.35)a	3.6 (0.41)a	6.2 (0.41)a
1	2	50	6.1 (2.92)a	16.2 (8.07)def	48.1 (25.72)abc	1.7 (0.34)a	3.1 (0.43)a	5.9 (0.47)a
1	4	100	5.9 (2.75)a	15.5 (7.63)bcde	45.3 (24.38)abc	1.9 (0.35)a	3.3 (0.42)a	5.9 (0.43)a
1	4	50	5.9 (2.94)a	16.4 (8.53)def	51.1 (28.79)bcd	1.8 (0.35)a	3.2 (0.44)a	6.0 (0.47)a
1	6	100	6.4 (2.65)a	15.5 (6.94)bcdef	41.1 (20.7)ab	2.2 (0.35)a	3.6 (0.41)a	6.2 (0.41)a
1	6	50	6.2 (3.22)a	18.1 (9.67)f	58.3 (34.26)cd	1.7 (0.35)a	3.1 (0.45)a	6.1 (0.49)a
1	9	100	5.1 (2.70)a	15.2 (8.31)abd	50.1 (29.96)abc	1.6 (0.34)a	2.8 (0.44)a	5.5 (0.48)a
1	9	50	5.2 (3.22)a	18.0 (11.21)cef	71.1 (47.18)d	1.7 (0.35)a	3.1 (0.45)a	6.1 (0.49)a

<sup>1</sup>Percent reduction data are days relative to the control of one corn, no weed competition, and 100% VWC.

<sup>2</sup>Means within the same column followed by the same lowercase letter are not statistically different ( $p \geq 0.1$ ).

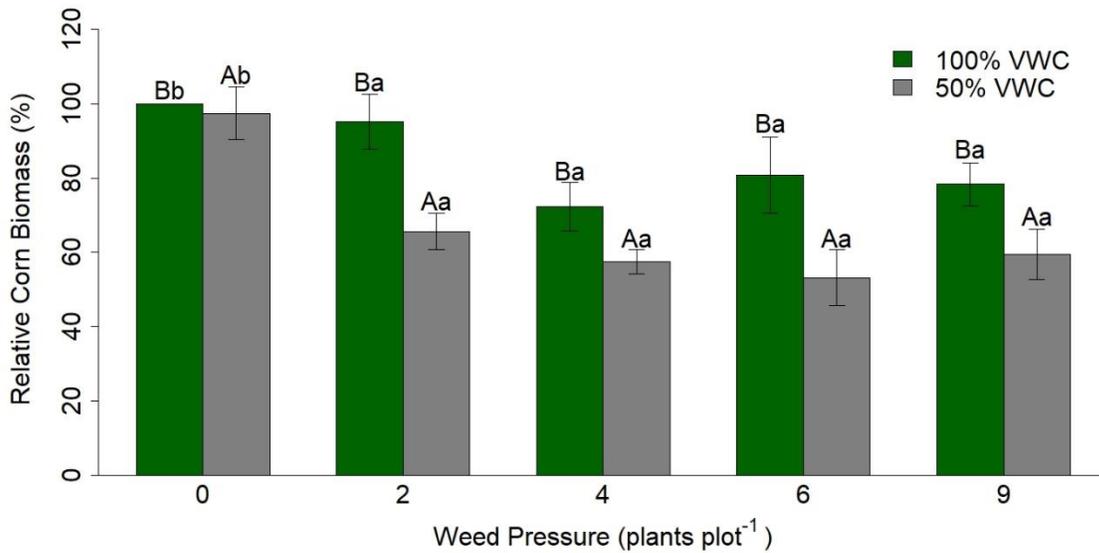


Figure 3.2. Mean (SE) relative drought tolerant corn biomass impacted by four weed pressures and two soil volumetric water content (VWC) levels in a greenhouse study. Bars labeled by the same capital letter are not statistically different for the main effect of VWC ( $p \geq 0.05$ ). Bars labeled by the same lowercase letter are not statistically different for the main effect of weed pressure ( $p \geq 0.05$ ). Biomass reduction data are relative to the control of one corn plant  $\text{pot}^{-1}$ , no weed competition, and 100% VWC.

Table 3.4. Mean (SE) drought tolerant corn photosynthetic response (Phi2, PhiNO, and PhiNPQ) to four weed pressures and two soil volumetric water content (VWC) levels at 14 and 21 days after water stress initiation in a greenhouse study.

Treatments <sup>1</sup>			Phi2 <sup>1</sup>		PhiNPQ <sup>1</sup>		PhiNO <sup>1</sup>	
Corn	Weed	VWC	14	21	14	21	14	21
Plant pot <sup>-1</sup>		%	Days					
1	0	100	0.60 (0.02)Aa <sup>2,3</sup>	0.53 (0.10)Ba <sup>2,3</sup>	0.24 (0.01)Aa <sup>2,3</sup>	0.31 (0.12)Ab <sup>2,3</sup>	0.16 (0.01)Aa <sup>2,3</sup>	0.16 (0.01)B <sup>2</sup>
1	0	50	0.58 (0.02)Aa	0.47 (0.07)Aa	0.26 (0.03)Aa	0.40 (0.09)Bb	0.16 (0.01)Aa	0.13 (0.03)A
1	2	100	0.61 (0.01)Aa	0.59 (0.01)Bab	0.23 (0.01)Aa	0.24 (0.01)Aab	0.17 (0.01)Aa	0.17 (0.02)B
1	2	50	0.59 (0.03)Aa	0.51 (0.03)Aab	0.25 (0.03)Aa	0.33 (0.04)Bab	0.16 (0.01)Aa	0.16 (0.02)A
1	4	100	0.56 (0.08)Aa	0.61 (0.03)Bb	0.27 (0.08)Aa	0.23 (0.03)Aa	0.17 (0.01)Aa	0.16 (0.00)B
1	4	50	0.58 (0.01)Aa	0.57 (0.03)Ab	0.26 (0.01)Aa	0.28 (0.03)Ba	0.16 (0.01)Aa	0.15 (0.00)A
1	6	100	0.60 (0.02)Aa	0.59 (0.03)Bb	0.23 (0.02)Aa	0.26 (0.04)Aa	0.17 (0.00)Aa	0.15 (0.01)B
1	6	50	0.58 (0.03)Aa	0.58 (0.05)Ab	0.27 (0.04)Aa	0.27 (0.05)Ba	0.16 (0.01)Aa	0.15 (0.01)A
1	9	100	0.58 (0.01)Aa	0.56 (0.06)Bab	0.25 (0.01)Aa	0.28 (0.05)Aab	0.16 (0.01)Aa	0.17 (0.01)B
1	9	50	0.58 (0.01)Aa	0.55 (0.03)Aab	0.26 (0.01)Aa	0.30 (0.03)Bab	0.16 (0.01)Aa	0.15 (0.00)A

<sup>1</sup>Abbreviations: VWC=percent soil volumetric water content, Phi2=quantum yield of Photosystem II, PhiNPQ=quantum yield of non-photochemical quenching, PhiNO=quantum yield of other unregulated losses.

<sup>2</sup>Means within the same column followed by the same capital letter are not significantly different ( $p \geq 0.1$ ) for the main effect of VWC.

<sup>3</sup>Means followed by the same lowercase letter are not significantly different ( $p \geq 0.1$ ) for the main effect of weed pressure.

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