QUANTIFYING POTENTIAL PRODUCTION RISKS OF TECHNOLOGIES NEARING COMMERCIALIZATION, MANAGEMENT PRACTICES FOR COTTON IRRIGATION, AND LEAF PUBESCENCE AND DEFOLIATION STRATEGY INFLUENCE ON COTTON DEFOLIATION AND FIBER QUALITY

by

SETH ANDREW BYRD

(Under the Direction of A. STANLEY CULPEPPER)

ABSTRACT

A variety of issues potentially impact cotton production and management of the crop, including new herbicide technologies, concerns over agricultural water use, and managing the crop to maximize yield and fiber quality. Multi-location trials were conducted in Georgia and across the cotton belt to evaluate the impact of sub-lethal rates of 2,4-D on cotton at various growth sages, the potential water savings benefits from a conservation tillage system utilizing a high biomass rolled rye cover crop system in cotton, and the influence of leaf pubescence and defoliation strategy on cotton fiber quality. Growth stages around the early bloom period were determined to be the most sensitive to sub-lethal rates of 2,4-D, primarily due to reductions in the number of bolls produced by damaged plants. Visual injury ratings and measurements of chlorophyll *a* fluorescence of cotton did not reflect yield loss resulting from 2,4-D exposure. The high biomass rolled rye conservation tillage system resulted in minimal benefits in soil moisture, cotton growth and development, and yield. In two of four locations, plant growth was negatively impacted by the conservation tillage system, while yield was lower in one location due to conservation tillage compared to conventional tillage. It appears that potential benefits gained from this system are highly dependent on environmental conditions, while detriments may be present in years when excessive rainfall occurs during the growing season. Multiple physiological measurements also illustrated minimal impact due to conservation tillage compared to conventional tillage. Varying water supply treatments were included, and typically benefits in cotton growth, development, and yield were observed in treatments that supplied the crop with the two greatest amounts of irrigation. Similar results were observed in physiological parameters, as leaf water potential and stomatal conductance were reduced when cotton received no supplemental irrigation compared to fully irrigated cotton. Minimizing leaf grade and trash content in ginned lint is one method to achieve maximum fiber quality. Trials determined that while a desiccating defoliation strategy can hamper cotton defoliation, leaf pubescence of cultivars is the primary driver behind the amount of leaf and trash content that remains in ginned lint.

INDEX WORDS: Cotton; 2,4-D; Enlist™; plant mapping; injury ratings; conservation tillage; cover crop; irrigation; leaf pubescence; defoliation; fiber quality.

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by

SETH ANDREW BYRD
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MS, University of Florida, 2012

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by

SETH ANDREW BYRD

Major Professor:	A. STANLEY CULPEPPER
Committee:	GUY D. COLLINS
	PHILLIP M. ROBERTS
	JOHN L. SNIDER
	JARED R. WHITAKER
	WESLEY M. PORTER

Electronic Version Approved:

Suzanne Barbour
Dean of the Graduate School
The University of Georgia
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DEDICATION

To Mom and Dad.

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CHAPTER 1

INTRODUCTION AND LITERATURE REVIEW

Introduction

Cotton production is facing a variety of changes and challenges that may shape the methods and practices used in future cotton production. The release of new technologies enabling the application of synthetic auxin herbicides, including 2,4-D resistant cotton, termed Enlist™ (Dow AgroSciences 9330 Zionsville Road, Indianapolis, IN 46268) and dicamba resistant cotton, termed Xtend™ (Monsanto Company 800 N. Lindbergh Blvd., St. Louis, MO 63167) will alter the way weeds are managed and cultivars are selected, however, there is a degree of risk involved with the adoption of these technologies. The release of Enlist™ and Xtend™ cotton will increase the amount of 2,4-D and dicamba applied in cotton producing areas, although the adoption of cultivars with this technology will not be immediately 100%. The presence of cultivars without tolerance to 2,4-D or dicamba, and the lack of cross-resistance between Enlist™ and Xtend™ cultivars in the dense areas of cotton production located throughout the United States, has heightened concerns about potential off-target movement of the synthetic auxin herbicides. Cotton is highly sensitive to synthetic auxin herbicides, 2,4-D specifically (Staten, 1946; Robinson and Fox, 1978), thus off-target movement or misapplication of these herbicides onto cultivars without this new herbicide resistance increases the potential for cotton injury and yield loss. 2,4-D has been a popular choice as a burndown (prior to planting) herbicide for decades, and numerous previous studies have observed the detrimental effects of 2,4-D drift on cotton development and yield when exposed during early stages of growth

(Sciumbato et al., 2004; Marple et al., 2007, 2008; Everitt and Keeling, 2009; and Johnson et al., 2012a). With the ability to use 2,4-D post-emergence in cotton over a long window of time, this technology creates the potential for cotton non-tolerant to 2,4-D to be exposed to the herbicide at a wide range of growth stages. Evaluating the response of cotton to sub-lethal drift or tank contamination rates of 2,4-D at various growth stages across the cotton belt could aid in mitigating yield loss and determining management practices to maximize returns when instances of 2,4-D exposure occurs in non-tolerant cotton.

Visual injury ratings have been used in the past to quantify the magnitude of damage resulting from off-target movement of herbicides on crops, specifically for the effects of 2,4-D on cotton which elicits very unique vegetative injury symptoms (Sciumbato et al., 2004; Marple et al., 2008; Everitt and Keeling, 2009). Due to the lack of consistency between visual injury ratings and yield loss in cotton exposed to 2,4-D (Johnson et al. 2012a), determining if an alternative exists to evaluating the magnitude of injury that corresponds to yield loss could serve a critical role for producers as 2,4-D usage and instances of off-target movement increases. Chlorophyll *a* fluorescence parameters have been reported in previous studies to detect the effects of PS I and PS II inhibiting herbicides (Haynes et al., 2000; Ralph, 2000; Eullaffroy and Vernet, 2003) but the effects of 2,4-D to agronomic crop species on these parameters appear to be unexplored. Decreased photosynthetic rates, damage to chloroplasts, and a reduction in stomatal aperture have been observed due to 2,4-D, but it is unclear if these will be reflected in measurements of chlorophyll *a* fluorescence (Grossman et al., 1996; Romero-Puertas, 2004; Grossman, 2010). Relatively new developments in fluorescence sensing allow for the ability to rapidly take a large amount of chlorophyll *a* fluorescence measurements over a short period of time using OJIP transients. This elevates OJIP transient measurement as a candidate for determining the potential of utilizing chlorophyll *a* fluorescence parameters to quantify 2,4-D injury and correlate the effects on fluorescence parameters to yield loss. If a strong relationship between fluorescence parameters and 2,4-D injury, and more importantly yield loss, can be documented then chlorophyll *a* fluorescence could become a tool for predicting the magnitude to damage and the associated yield loss expected from cotton exposed to sub-lethal rates of 2,4-D.

Another critical issue facing cotton production, and agriculture in general, is water use. Recent federal (EPA, 2015) and state (GDNR – EPD, 2011; GDNR-EPD, 2012) regulations have increased concerns regarding agricultural water use. Coupled with climate variability including unpredictable rainfall and episodic droughts, increasing efficiency of water use from both an irrigated and non-irrigated production environments will be critical to maintaining economically sound production moving forward. Efficient water use on cotton is of particular concern in Georgia as it is the largest row crop in the stage on an acreage basis, and is the crop most commonly grown without irrigation. Multiple studies have documented the negative impacts of water deficit on the growth and yield of cotton (Gerik et al., 1996; Pringle and Martin, 2003; Pettigrew, 2004a; Whitaker et al., 2008; Gwathmey et al., 2011), thus increasing the efficiency of the utilization of water, through either irrigation or rainfall, will be key to mitigating periods of water deficit and avoiding the detrimental effects of water stress. One method that has been successful in the past at increasing soil moisture, thus mitigating stress from periods of water deficit is conservation tillage with the employment of cover crops (Blevins et al., 1971; Gantzer and Blake, 1978; Mills et al., 1988; Dao, 1993; Daniel et al., 1999b). Additionally, infiltration and retention of water in the soil profile have been observed to increase with the use of cover crops compared to conventional tillage (Dao, 1993; Lascano et al., 1994; Bruce et al., 1995; Raper et al., 2000) The utilization of a cover crop in cotton could not only aid dryland producers in retaining soil moisture for longer periods of time between rainfall events, but could also allow for more efficient use and a reduction in application rates of irrigation.

While the end result of water stress in crops is typically a reduction in yield, the initial effect occurs on physiological processes impacted by plant water status. Reductions in leaf water potential, stomatal conductance and photosynthetic rates have been observed due to water deficit in cotton (Radin, 1984; Turner et al., 1986; Medrano et al., 2002; Flexas et al., 2002; Snider et al., 2014). These measurements or processes are inherently linked, as reductions in photosynthetic rates have been found to be directly related to reductions in stomatal conductance (Medrano et al., 2002; Flexas et al., 2002; Snider et al., 2014), which in turn is directly impacted by leaf water potential (Radin, 1984; Turner et al., 1986; Snider et al., 2014). The benefits of increased soil moisture (Blevins et al., 1971; Gantzer and Blake, 1978; Mills et al., 1988; Dao, 1993; Daniel et al., 1999b) and soil water retention (Dao, 1993; Lascano et al., 1994; Bruce et al., 1995; Raper et al., 2000) have been documented when comparing crops grown under conservation tillage to conventional tillage systems. While numerous previous studies have quantified the effects on plant growth and yield from the utilization of cover crops, the physiological impacts have been evaluated to a lesser extent, particularly in cotton under varying water availability. If increased water availability could be achieved through the implementation of a conservation tillage system utilizing cover crops to a degree that increases physiological function compared to crops grown under conventional tillage, further understanding of the potential benefits of this system could be gained, particularly under water deficit conditions.

Beyond maximizing yield, achieving high fiber quality is a goal for cotton producers, though concerns over fiber quality have recently increased (Shurley and Collins, 2013). One of the primary drivers behind optimal fiber quality is proper use of harvest-aid or defoliation.

When optimal defoliation takes place, the live, green plant material is removed from the plant, minimizing the amount of dead plant material that ends up in harvested seedcotton, maintaining the quality by reducing the amount of plant material, or trash, in the ginned lint (Colwick et al., 1984; Brecke et al., 2001; Valco and Snipes, 2001). Two factors that influence the effectiveness of defoliation, and in turn the amount trash that is contained in ginned lint, are defoliation strategy and leaf pubescence characteristics of cultivars. Defoliants are one category of harvest-aids that lead to the removal of leaves by forming an abscission layer at the base of the petiole on live, healthy leaves. Defoliants are commonly recommended to be included in harvest-aid mixtures for cotton (Collins et al., 2015). Desiccants, another form of harvest-aid, result in rapid death of plant tissues which prevents the formation of an abscission layer and often results in leaf stick, or dead plant material being left on the plant (Stahler, 1953; McMeans et al., 1966; Bovey and Miller, 1968; Brecke et al., 2001; Shaw, 2002). This often leads to increased trash in the ginned lint due to the dead plant material stuck to the plant being harvested along with the seedcotton (Shaw, 2002). Leaf pubescence characteristics of cultivars also influences fiber quality, as cultivars with increased leaf pubescence, or hairy cultivars, often have reduced fiber quality due to increased trash levels in ginned lint resulting from the hairier leaves being entangled in the lint of open bolls upon defoliation (Ramey, 1962; Smith, 1964; Wanjura et al., 1976; Novick et al., 1991). Because increased trash levels could result in reduced lint value, it is critical that producers are aware of how defoliation strategies and cultivar characteristics influence fiber quality characteristics.

Literature Review

Impact of Sub-Lethal Rates of 2,4-D at Various Growth Stages of Cotton. The advent of 2,4-D resistant cotton, termed Enlist™ by Dow and dicamba resistant cotton by Monsanto branded

Xtend™ (Johnson et al., 2012b) is being commercialized and made available to cotton growers in the Southeastern US in the near future. While acceptance of these new technologies are expected among producers, many may continue to plant less modern cultivars due to costs or anxiety related to potential drift of respective herbicides onto non-target crops or tank cleaning issues. The release and adoption of these cultivars by growers will most certainly increase the interface between tolerant and non-tolerant crops in adjacent fields or by neighboring growers in a close proximity.

The synthetic hormone 2,4-dichlorophenoxyacetic acid (2,4-D) was discovered as a plant growth regulator in the mid-1940s and was available on the commercial market as an herbicide by 1945 (Peterson, 1967). The chemical was widely adopted and utilized in the agricultural market, and helped facilitate the more than $267,000,000 increase in spending on chemical weed control between 1940 and 1962 (Peterson, 1967). The chemical 2,4-D behaves as an auxin within the plant, with a very high activity when compared to other auxins, and it is this high auxin activity that results in the death of the plant (van Overbeek et al., 1950). The natural production of metabolites is greatly increased in response to 2,4-D, while the high auxin activity also increases sensitivity of plant tissue to damage by these metabolites (van Overbeek et al., 1950). The production of coumarin derivative compounds, which are phytotoxic metabolites, are increased in response to 2,4-D culminating in the death of the plant (van Overbeek et al., 1950). Broadleaf plant species are more susceptible to phytoxic compounds than grass species resulting in the selective action of 2,4-D (Hamner et al., 1950; van Overbeek et al., 1950). Registered in 1967, dicamba is another herbicide that behaves like 2,4-D, mimicking auxin to cause abnormal cell division leading to the death of the plant (Cox, 1994). Once absorbed by the plant, dicamba is translocated throughout the plant and accumulates in new young plant tissue, affecting meristematic tissue (Cox, 1994). The symptoms of 2,4-D and dicamba injury are similar, including malformation of leaves and stem (epinasty, cupping, curling, etc.) and excessive growth (Al-Khatib and Peterson, 1999; NPIC, 2002).

The herbicide 2,4-D was found to be very effective for weed control as early as the mid-1940’s (Anderson and Wolf, 1947), and it has been established as one of the most effective herbicides for control of broadleaf weeds for both burndown (Fawcett and Slife, 1978) and for weed management programs in corn and other grass species cropping systems (Triplett Jr. and Lytle, 1972). Control of troublesome weeds in turf and forage crops have been reported with 2,4-D alone (Kohler et al., 2004; Schnick and Boland, 2004; Hutto et al., 2007) as well as with 2,4-D in a mixture with other herbicides (Beeler et al., 2004), including mixtures with dicamba (Hutto et al., 2007). Improved weed control has been observed with multiple applications of 2,4-D compared to a single application (Hutto et al., 2004).

Success has also been reported for control of several problematic weeds in row crop production in studies utilizing 2,4-D and dicamba. Control of palmer amaranth, one of the most problematic weeds in southeastern cotton production, has been observed with 2,4-D (Jha and Norsworthy, 2012) and dicamba (Doherty et al., 2010; Jha and Norsworthy, 2012) applications. Previous studies have reported 2,4-D to be successful in the control of horseweed (Kruger et al., 2008; Kruger et al., 2010), sicklepod (Lancaster et al., 2005), giant ragweed, common waterhemp, common lambsquarters, velvetleaf (Robinson et al., 2012), red morningglory (Siebert et al., 2004), hairy vetch, crimson clover (White and Worsham, 1990), cocklebur, smartweed, and jimson weed (Williams et al., 1960), while dicamba has also been found to provide control of kochia (Nandula and Manthey, 2002) . Other researchers have found that both 2,4-D and dicamba provided control of horseweed and Russian thistle (Everitt and Keeling, 2007). Mixtures of 2,4-D with other herbicides have also been found to be effective in managing challenging weed species. Mixtures including 2,4-D have been reported to control several sub-species of morningglory (Culpepper et al., 2001) as well as horseweed (Kruger et al., 2010), giant ragweed, common waterhemp, common lambsquarters, velvetleaf (Robinson et al., 2012), hairy vetch and crimson clover (White and Worsham, 1990). Siebert et al. (2004) observed successful control of red morningglory with a mixture of dicamba and 2,4-D, while White and Worsham (1990) also reported control of hairy vetch and crimson clover with a mixture of paraquat and 2,4-D. Several of the studies mentioned above also investigated the effect of 2,4-D and dicamba on weed seed production and reported that either 2,4-D (Lancaster et al., 2005; Kruger et al., 2010; Jha and Norsworthy, 2012) or dicamba (Jha and Norsworthy, 2012) reduced the production of weed seeds.

Visible ratings of injury on weeds subjected to 2,4-D or dicamba has been reported to be significant between seven and 42 days after application (Culpepper et al., 2001; Nandula and Manthey, 2002; Siebert et al., 2004; Kruger et al, 2008; Doherty et al., 2010; Kruger et al., 2010; Jha and Norsworthy, 2012; Robinson et al., 2012), compared to weeds with no herbicides applied, with the typical timing of the most severe symptoms often occurring 28 days after treatment (Kruger et al, 2008; Jha and Norsworthy, 2012; Robinson et al., 2012). The typical visual symptoms of 2,4-D and dicamba injury is abnormal growth (Fawcett and Slife, 1978) including epinasty, leaf cupping, and the senescence of the leaves and petioles (Kohler et al., 2004) resulting in the reduction in plant dry weight (Kruger et al., 2010; Robinson et al., 2012).

In the past decade a shift has occurred in weed populations due to the development of glyphosate resistance in certain weed species (Culpepper, 2006; Webster and Sosnoskie, 2010). Widespread use of glyphosate in cropping systems, as well as multiple applications of the herbicide throughout the growing season, increased selection pressure on weed populations and led to the development of weed species resistant or tolerant to glyphosate (Mueller et al., 2005). Specifically, cotton producers in Georgia have witnessed an increase in the number of glyphosate resistant weeds present, with GR Palmer amaranth first discovered in Georgia in 2005 (Culpepper et al., 2006), being the most problematic weed in terms of management and economic effect (Rowland et al., 1999; Smith et al., 2000; Morgan et al., 2001). In 2007, 1.6 million pounds of glyphosate was used on cotton in Georgia, while glyphosate was applied on 88% of the cotton land area grown in Georgia in 2010 (USDA – NASS, 2013). The introduction of 2,4-D and dicamba resistant cotton cultivars may improve control of GR weeds, as both herbicides have been found to provide control of Palmer amaranth in previous studies (Doherty et al., 2010; Jha and Norsworthy, 2012). These new technologies may also combat the development of additional weed species developing tolerance to glyphosate as well as glufosinate, another widely used herbicide in cotton, as repeated applications of the same herbicidal mode of action hastens the development of weed resistance (Jasieniuk et al., 1996). The addition of 2,4-D and dicamba resistance in cotton will provide producers an additional mode of action not previously available for post emergent applications (Craigmyle et al., 2013) and prolong the effectiveness of currently used herbicides.

Reports of injury to cotton due to 2,4-D also date back to the 1940's, as Slaten (1946) reported injury to the terminal growing points of cotton exposed to 2,4-D drift from adjacent fields, with injury occurring more than 18 meters from the field border. It was also reported that the amount of injury sustained by cotton subjected to some form of 2,4-D exposure varied depending to the level, or concentration, of the herbicide solution (Slaten, 1946). Early studies have also observed the malformation of leaves (McIlrath and Ergle, 1952), death of cotton squares and flowers after exposure to 2,4-D, as well as suppressed vegetative growth and a delay in new boll development of approximately 60 days following applications of 2,4-D (McIlrath et al, 1951).

While previous studies have reported that 2,4-D and dicamba applications to non-tolerant cotton prior to emergence result in injury or reduced stands (Baker, 1993; York et al., 2004; Everitt and Keeling, 2007), the focus of this study is on the effect of post-emergence applications. Applications of 2,4-D and dicamba to cotton post emergence has been observed to result in visual injury in multiple previous studies (Sciumbato et al., 2004; Marple et al., 2007). Injury to cotton from either 2,4-D or dicamba exposure have been reported to be more severe to plants in early, vegetative growth stages compared to applications made during later stages of growth, with increasing injury symptoms as herbicide rates increase (Marple et al., 2008; Everitt and Keeling, 2009). Symptoms of 2,4-D or dicamba injury to cotton are visually similar, including leaf cupping, strapping, and cholorsis, twisting of petioles, stem epinasty, stunting of growth, and loss of apical dominance (Marple et al., 2008; Everitt and Keeling, 2009; Johnson et al., 2012a). Visual ratings of leaf and stem malformations are what typically have been used to determine severity of injury in cotton exposed to 2,4-D and dicamba (Sciumbato et al., 2004; Marple et al., 2008; Everitt and Keeling, 2009). Exposure of cotton to 2,4-D or dicamba during early stages of growth have also been observed to have a negative impact on yield (Marple et al., 2008; Johnson et al., 2012a). The greatest visual injury due to 2,4-D application in cotton has been determined to be present 28 days after the application (Marple et al., 2007), while Marple et al. (2008) reported that visual estimates of injury 28 days after application of 2,4-D were most correlated to yield loss. This closely correlates to findings of the timing of visual injury to target weeds in the aforementioned studies. Foliar malformation due to dicamba injury has been observed to increase during the first three weeks after application on new growth, with a reduction in flowering reported when deformed leaves are present (Hamilton and Arle, 1979). Recovery from dicamba injury has typically been observed to begin at approximately 28 days after application (Hamilton and Arle, 1979; Marple et al., 2007).

Application of either 2,4-D or dicamba to cotton has been found to result in decreased yields compared to non-treated cotton (Smith and Wiese, 1972; Hamilton and Arle, 1979; Marple et al., 2007; Everitt and Keeling, 2009; Johnson et al., 2012a). Dicamba applied to cotton during pre-bloom stages resulted in reduced micronaire (Smith and Wiese, 1972), while Hamilton and Arle (1979) reported defoliation effectiveness was reduced when dicamba was applied to cotton between 12 and 21 weeks after emergence. Micronaire reductions were reported when 2,4-D was applied at either the pre-square, squaring, or blooming growth stages (Smith and Wiese, 1972). Cotton treated with 2,4-D had fewer flowers and greater flower abortion than non-treated cotton (Marple et al., 2007), with fiber elongation also reduced in 2,4-D treated cotton (Marple et al., 2008).

Several of the aforementioned studies modeled the 2,4-D or dicamba treatments to simulate drift using fractions of recommended rates to represent off-site movement into cotton fields (Marple et al., 2007, 2008; Everitt and Keeling, 2009; Johnson et al., 2012). Physical drift is one of the main concerns regarding the release of 2,4-D and dicamba resistant cotton cultivars, with new cultivars of resistant cotton being located adjacent to fields of non-resistant cotton. The severity of drift, or off-site movement of the herbicide, is a function of the physical condition of the field, including crop height, as well as equipment parameters such as the droplet size, nozzle height, and speed of movement, and atmospheric or ambient factors such as wind speed, temperature, and humidity (Hanks et al., 1995; Holterman et al., 1997). Smith and Wiese

(1972) determined that cotton three meters away from plants applied with 2,4-D sustained 37 – 50 percent damage, while cotton 12 meters away sustained 7 – 20 percent damage. Dicamba drift had less severe results, but still resulted in 15 – 30 percent damage present on cotton 3 meters away, and 0-7 percent damage on cotton 12 meters away (Smith and Wiese, 1972). In Australia, issues with 2,4-D drift onto cotton from neighboring wheat fields has been an issue, due in large part to the high volatility of 2,4-D formulations used which enables the herbicide to drift over several kilometers (Cousins et al., 1991).

New cotton cultivars with 2,4-D or dicamba resistance will offer growers a simplified weed control program and could perhaps reduce the number of applications and different herbicides used in cotton production (Johnson et al., 2012b). Cotton cultivars that expressed resistance to 2,4-D were originally developed in the late 1980s and early 1990s and were intended to provide drift protection of these sensitive crops from 2,4-D applications in adjacent fields (Laurent et al., 2000). The synthetic enzymes that kill weeds or plants sensitive to 2,4-D or dicamba are metabolized in the new resistant crop cultivars, allowing for direct application of the herbicide without damage (Laurent et al., 2000). Both the 2,4-D and dicamba formulations that will be used on the resistant cotton will include enhancements that are touted to reduce the volatility and drift of the herbicide (Johnson et al., 2012b). Weeds developing resistance to herbicides is another issue that has arisen in the past with crops engineered with herbicide resistance, such as Roundup Ready® technology (Johnson et al., 2012b). This situation is anticipated to be avoided with 2,4-D and dicamba, due to their already extensive and long term use in crops with little resistance in weeds developing, and their superb control of several weed species that have developed Roundup resistance (Johnson et al., 2012b). These new technologies will also offer a previously unavailable mode of action to combat weeds that are resistance to currently utilized herbicides in cotton production, as well as inhibit the development of resistances forming in additional weed populations.

Potential for Utilizing Chlorophyll a Fluorescence to Detect Injury and Predict Yield Loss from Sub-Lethal Rates of 2,4-D on Cotton. The synthetic auxin herbicide 2,4-dichlorophenoxyacetic acid (2,4-D) has been one of the most widely used agrochemicals since the 1940’s (Peterson, 1967; Grover et al., 1972). The ability to control broadleaf weeds in cereal crops, as well as the inexpensiveness of the herbicide, made 2,4-D and other synthetic auxin compounds, one of the first classes of herbicides produced for agricultural use and a popular choice among growers (Peterson, 1967; Grover et al., 1972; Bayley et al., 1992; Grossmann, 2000; Mithila et al., 2011). However, the high volatility of several widely used formulations of 2,4-D have raised issues of off-target movement of the herbicide, resulting in drift damage to neighboring, non-target crop species dating back to the 1950’s (Robinson and Fox, 1978; Bayley et al., 1992). Cotton is regarded as one of the most sensitive crop species in terms of susceptibility to damage from 2,4-D drift, with extensive losses occurring due to off-target movement of 2,4-D onto cotton (Robinson and Fox, 1978; Bayley et al., 1992; Lyon et al., 1993). Due to the widespread use of 2,4-D and other synthetic auxin herbicides, and the sensitivity of cotton to these herbicides, the development of cotton resistant to 2,4-D has been a focus of research since the early 1990’s (Bayley et al., 1992; Lyon et al., 1993). The primary objective of these previous studies was to reduce or eliminate damage to cotton from drift of 2,4-D from applications on nearby cereal crop fields (Lyon et al., 1993; Laurent et al., 2000). However, with the scheduled upcoming release of 2,4-D resistant cotton (Enlist™) from Dow AgroSciences (Dow AgroSciences, Indianapolis, IN), as well as dicamba resistant cotton (Xtend™) from Monsanto Company (Monsanto Company, Saint Louis, MO), post emergence applications of synthetic auxin herbicides to cotton will be made possible (Johnson et al., 2012b). The release of these technologies will increase the interface between tolerant and non-tolerant cotton as well as increase the potential for drift injury to cotton from such herbicide applications.

While this advancement in engineering for herbicide tolerance could provide a broader spectrum of weed control, as well as another mode-of-action (MOA) option for post emergence applications for growers (Johnson et al., 2012b), concerns exist about what effect the adoption of these new technologies will have on non-tolerant cotton. It is likely that a significant portion of cotton acreage will continue to contain cultivars without resistance to 2,4-D or dicamba. Specifically, there are concerns of drift damage from post emergent applications to resistant cotton cultivars located in close proximity to fields of cotton that are susceptible to 2,4-D or dicamba damage. In the southeastern United States, and Georgia in particular, multiple scenarios exist in regards to the cropping system and management practices utilized by individual producers, including various planting dates, cultivars, and chemical application practices and schedules. This creates the potential for various growth stages of cotton simultaneously exposed to drift from 2,4-D or dicamba applications on neighboring fields of resistant cotton cultivars. The development of a plant based measurement that can be utilized to rapidly determine the injury level sustained by cotton plants damaged by 2,4-D or dicamba drift, indexed to yield reductions and growth stage parameters, could be a key to quantifying and correlating plant injury at various growth stages to yield loss.

Chlorophyll a fluorescence-based measurements to quantify herbicide injury have been used in the past for a variety of plant species and herbicide combinations. Because chlorophyll fluorescence is a rapid method (1-2 seconds per sample) for quantifying photosynthetic efficiency in plants, fluorescence is a candidate not only for the determination of 2,4-D and dicamba drift injury to cotton, but fluorescence measurements could also potentially be correlated with yield loss to provide growth-stage specific fluorescence thresholds at which a given level of yield loss could be expected.

Auxin is a vital plant hormone, containing indole-3-acetic acid (IAA), and is a critical requirement for plant growth and development (Woodward and Bartel, 2005; Grossmann, 2010). As the concentration of auxin increases inside cells, it elicits a change in the developmental growth of plants (Vanneste and Friml, 2009). Auxins regulate the elongation and division of cells, and are known to be the key to differential development in plants, being required for the development of vascular tissue, lateral roots, floral meristems, and leaves among other structures (De Smet et al., 2003; Woodward and Bartel, 2005; Grossmann, 2010). Auxin is typically biosynthesized in the apical regions of shoot tissue, but can be transported cell to cell in response to environmental or internal plant signals (Woodward and Bartel, 2005; Vanneste and Friml, 2009).

Phenoxy herbicides, such as 2,4-D, are part of a class of herbicides typically referred to as synthetic auxins. Growth, through cell division and elongation, is a typical plant response to auxin in low cellular concentrations (Grossmann, 2000). This increase in auxin concentration is followed by an increase in the expression of genes that dissipate the action of auxin and IAA, which regulates the rate of cell growth in response to auxin (Mithila et al., 2011). Due to structural similarities to natural auxin compounds, synthetic auxin herbicides imitate natural auxins in the plant, resulting in the same plant growth responses as are present with an increase in natural auxin concentrations (Grossmann, 2010; Mithila et al., 2011). When auxin is present in high concentrations, irregular plant growth can occur, which is typically counteracted by the plant internally through the synthesis compounds to deter these effects and correct the hormonal imbalance within the cell (Grossman, 2000; Hansen and Grossmann, 2000; Grossmann, 2010). With the application of herbicides such as 2,4-D, the synthetic auxin compounds bind to the auxin binding sites and are transported into the cells by carrier proteins that serve this function for natural auxin compounds as well (Mithila et al., 2011). At this point, in the case of a natural compound, the increase in cellular auxin concentration would result in the expression of genes that suppress auxin, thus regulating the growth stimulus. However 2,4-D is not suppressed by the expression of these genes and the repressive compounds produced through the expression of these genes (Mithila et al., 2011). This results in a build-up of synthetic auxin in the plant and a disproportionate level of auxin in the plant tissue, which subsequently results in the signaling of other plant hormones and compounds (Grossmann et al., 2010). Previous research has determined that this synthesis of other plant compounds, in response to increased auxin levels, is the key to the herbicidal properties of 2,4-D and other synthetic auxin herbicides (Grossmann, 2000; Hansen and Grossmann, 2000; Grossmann, 2010; Mithila et al., 2011).

The effects and visual symptoms of 2,4-D damage on plants have been well documented in previous research. Grossmann (2010) describes the processes and stages through which synthetic auxin herbicides lead to plant death. The initial stage takes place in shoot tissues, in response to increased auxin (or synthetic auxin in this case), where induction of 1-aminocyclopropane-1-carboxylic acid (ACC) synthase occurs resulting in the synthesis of ethylene (Grossmann, 2010). A period of abnormal growth follows, including leaf and stem epinasty that are the characteristic visual injury symptoms of 2,4-D and other synthetic auxin herbicides (Al-Khatib and Peterson, 1999; Grossmann et al., 2001; Grossmann, 2010). Subsequently, levels of abscisic acid (ABA) increase, causing suppressed root and stem growth, as well as a reduction in stomata aperture, impeding the assimilation of carbon (Grossmann, 2010). It is known that with reduced stomata aperture, CO_2 diffusion to the carboxylation site is inhibited, thereby decreasing photosynthetic rates (Vaadia et al., 1961; Hsiao and Acevedo, 1974; Grossmann et al., 2001; Flexas et al., 2004; Saibo et al., 2008; Lawlor and Tezara, 2009). These conditions lead to senescence, deterioration of plant tissues including the vascular tissue, necrosis, and ultimately plant death (Grossmann, 2000; Grossmann, 2010).

Ethylene is one of the compounds reported to be synthesized in response to elevated levels of auxin or IAA in plant tissues (Burg and Burg, 1966; Abel et al., 1995). Ethylene has suppressive effects on plant growth (Burg and Burg, 1966; Suttle, 1988) and can alter both the synthesis and cellular transport of auxin (Suttle, 1988; Vanneste and Friml, 2009). This increase in ethylene synthesis resulting from ACC synthase in response to elevated auxin levels is also observed in plants receiving applications of synthetic auxin herbicides (Morgan and Hall, 1962; Grossman, 2000; Hansen and Grossman, 2000; Grossmann, 2010). Ethylene stimulates cell growth in a lateral orientation which gives rise to the visual malformation symptoms of synthetic auxin herbicide injury (Grossmann, 2010). The increased synthesis of ethylene in cotton plants has been observed with applications of sub-lethal doses of 2,4-D, with the production of ethylene increasing with increasing application amounts of 2,4-D (Morgan and Hall, 1962). A co-product of the process that synthesizes ethylene, in conjunction with ACC synthase, is cyanide, and an increase in cyanide levels has been observed in plants treated with 2,4-D (Tittle et al., 1990; Grossmann, 2000). Increases in cyanide levels have been found to be highly correlated with increases of ethylene synthesis in soybeans exposed to 2,4-D (Tittle et al., 1990). Tittle et al. (1990) reported marked increases in both ethylene and cyanide levels six hours after exposure to 2,4-D in soybeans. Cyanide, present at toxic levels, is believed to be a primary cause of the tissue deterioration found in the latter symptom phases of plants experiencing lethal doses of 2,4-D (Grossmann, 2000).

The application of synthetic auxin results in the increase of another component critical to their role as an herbicide, ABA. The hormone ABA acts as a growth regulator in plants, and exposure to ABA has been reported to inhibit the growth of lateral roots, and is thought to act as an auxin suppressor (De Smet et al., 2003). Increases in ABA levels have also been reported to occur in response to a rise in natural auxin, or IAA, concentrations in plants (Dunlap and Robacker, 1990). Similar observations regarding increased ABA levels have been reported when plants are exposed to synthetic auxin herbicides (Grossmann et al., 1996; Grossmann, 2000; Hansen and Grossmann, 2000; Grossmann et al., 2001; Grossmann, 2010). Typically thought of as drought response signal, ABA concentration has been observed to increase in the shoot tissue of a plant after application of 2,4-D, causing reduced stomatal aperture (Grossmann, 2000; Grossmann et al., 2001; Grossmann, 2010). As a result, carbon assimilation is decreased, leading to reduced photosynthetic rates and inhibition of plant growth (Grossmann et al., 1996; Grossmann, 2000; Hansen and Grossmann, 2000; Grossmann et al., 2001; Grossmann, 2010). A further effect of increased ABA in response to synthetic auxin herbicides is an increase in the presence of reactive oxygen species, which has been reported to be another key factor in the herbicidal properties of 2,4-D that lead to plant death (Grossmann et al., 2001; Romero-Puertas et al., 2004; Grossmann, 2010).

Exposure of susceptible plants to synthetic auxin herbicides, such as 2,4-D, results in an increased concentration of auxin, or auxin-imitators, compared to natural auxin levels, which stimulates the synthesis of multiple plant compounds. While this would typically lead to a suppression of auxin levels, in the case of synthetic auxin compounds the synthesis and accumulation of ethylene, cyanide, ABA and their co-products at toxic levels eventually results in unregulated growth, inhibition of physiological processes, and plant death. In the case of cotton, exposure to sub-lethal, or simulated drift rates of 2,4-D resulted in leaf and stem malformation (Marple et al., 2007, 2008; Everitt and Keeling, 2009; Johnson et al., 2012a) and the abortion of fruiting structures (Marple et al., 2007).

The term drift is widely used to describe off site movement of a spray application, and typically refers to a pesticide formulation. This off-target movement can lead to injury of non-target crops when herbicide drift occurs from applications on surrounding fields, and can have a severe economic impact (Baetens et al., 2009). The physical components of the field, including crop height, as well as equipment parameters such as the droplet size, nozzle height, and speed of movement, and atmospheric or ambient factors such as wind speed, temperature, and humidity all influence the severity of drift (Hanks et al., 1995; Holterman et al., 1997). As an agricultural herbicide, 2,4-D has been widely used for over 60 years, and damage from drift has been an issue since the 1950's (Robinson and Fox, 1978). Renne and Wolf (1979) reported that cool, humid conditions suppress the drift potential of 2,4-D by reducing evaporation, and increasing the retention, of spray droplets on vegetative surfaces.

Certain formulations of 2,4-D have been reported to be highly susceptible to drift during application, specifically the formulations classified as having greater volatility (Grover et al., 1972; Farwell et al., 1976; Robison and Fox, 1978). In terms of the distance from the site of application, 2,4-D drift can travel several kilometers, depending on existing weather patterns (Farwell et al., 1976), and Robinson and Fox (1978) determined that 2,4-D had travelled greater than 32 km in a study conducted in Washington. Even when drift results in exposure to sub-lethal doses, such as those expected to be present in plants exposed to drift of 2,4-D, the herbicide can have a significant effect. Cotton in particular is known to be one of the most sensitive crops in regard to injury from 2,4-D exposure (Robinson and Fox, 1978). Smith and Wiese (1972) reported that cotton three meters away from a targeted application of 2,4-D sustained 37 – 50% damage, while cotton 12 meters away sustained 7 – 20% damage. Staten (1946) reported cotton injury over 18 meters from the edge of plots sprayed with 2,4-D with wind speeds of approximately 21 km per hour and a nozzle height of less than one meter above the ground. Spray drift in this study was observed to extend greater than 21 meters from the site of application (Staten, 1946). Sub-lethal rates of 2,4-D have also been observed to cause severe growth and yield effects on a variety of vegetable crops (Hemphill and Montgomery, 1981).

Droplet size is a key component that determines the potential for drift to occur, with smaller droplets more prone to off-target movement (Hanks, 1995). Droplet size of 2,4-D can also determine the severity of injury once the droplet is deposited on a susceptible, or non-target crop species. In a study involving sunflower, McKinlay et al. (1972) observed greater damage from the smallest (100 μ) droplet size of 2,4-D applied. It was determined that larger droplet sizes (200 μ and 400 μ) would increase the severity of local cell necrosis in close proximity to the site of droplet deposition, while the integrity of cells adjacent to the location of smaller droplets would be maintained so that translocation of the herbicide could still occur (McKinlay et al., 1972). McKinlay et al. (1972) stated that three and six times as much active ingredient would need to be applied when using 200 μ and 400 μ droplet sizes, respectively, to equal the damage generated from 100 μ droplets. While being more prone to drift and more successful at creating conditions in which translocation can result, there is another component of small droplets that increase the detrimental impacts of drift. Maybank et al. (1978) reported that the density of off-target droplets has the potential to exceed the density of droplets at the site of application, depending on the droplet size and various environmental conditions, particularly with small spray droplets. These properties make increasing droplet size, while decreasing the amount of small droplets produced key factors to suppressing the drift fraction of pesticide applications (Maybank et al., 1978).

Due to drift issues and strong herbicidal activity on dicot crop species, injury due to drift of 2,4-D and other synthetic auxin herbicides has been an area of concern and a focus of numerous studies for the past 60 years. With the impending release of 2,4-D and dicamba tolerant cotton varieties, there is heightened concerns in regions of dense cotton production, such as south Georgia, where situations could exist in which new varieties, tolerant to these herbicides will be grown adjacent to areas of cultivars without resistance to 2,4-D or dicamba. This will inherently lead to applications of either herbicide in close proximity to fields containing susceptible cotton, generating the potential for drift injury.

Previous studies that have quantified the injury to cotton through simulated drift of 2,4-D have relied on visual observations of symptoms to rate the severity of plant injury (Smith and Wiese, 1972; Sciumbato et al., 2004; Marple et al., 2007, 2008; Everitt and Keeling, 2009; Johnson et al., 2012a). In studies that included cotton response to simulated drift of 2,4-D and dicamba, drift of dicamba led to less severe injury symptoms and yield reductions than 2,4-D (Smith and Wiese, 1972; Marple et al., 2007, 2008; Everitt and Keeling, 2009; Johnson et al., 2012a).   Previous work has also determined that the severity of visual injury symptoms and yield reduction in cotton is increased when plants are exposed to 2,4-D or dicamba early in the season, prior to bloom, compared to exposure during later growth stages (Marple et al., 2008; Everitt and Keeling, 2009; Johnson et al., 2012a;). While visual observation of injury symptoms is a common practice in determining the severity of injury to cotton exposed to drift of 2,4-D and other synthetic auxin herbicides, Johnson et al. (2012a) reported that depending on the timing of the visual ratings in regard to the timing of exposure of cotton to the herbicide drift, these visual estimates do not always consistently correlate with drift injury-induced yield losses. Hickman et al. (1991) evaluated the use of infrared images and video from an airplane flown over a cotton field that had received dicamba applications to determine crop injury and relate it to visual injury ratings. However, it was determined that the use of the still photographs and video, as well as reflectance measurements taken with a handheld meter, detected injury on between 35.7 - 47.5% of the total affected area as defined by visual determination (Hickman et al., 1991) suggesting that utilizing these method for injury detection is even less accurate than visual estimations. The use of aerial imagery, even if effective in determining injury sustained from drift of dicamba or 2,4-D, requires specialized equipment and is most likely too costly to be implemented in site by site evaluations.

Chlorophyll *a* fluorescence parameters have been measured in past studies for detection of changes in photosynthetic efficiency in plant species exposed to a variety of herbicides. In previous research, two parameters in particular, F_v/F_m and ϕ_{EO} (or ϕ_{PSII}) have been utilized to detect herbicide injury or damage to photosynthetic function. F_v/F_m is the maximum quantum yield of photosystem II and is a measure of potential efficiency with which photosystem II converts light energy into photosynthetic electron transport (Maxwell and Johnson, 2000). The ϕ_{EO} value is described as the quantum yield of electron transport (Strasser et al., 2004) and the probability of an electron entering the electron transport chain from photosystem II (Srivastava et al., 1998; Strasser et al., 2000). In algae, changes in the range of fluorescence emission spectra, which are reflective of photosystem I and II activity, were reported with exposure to the photosystem II inhibiting herbicides atrazine, diuron, metribuzin, and terbuthylazine (Eullaffroy and Vernet, 2003). Actual and quantum yield measurements of fluorescence in species of aquatic grass have been observed to decline with exposure to the herbicide diuron (Haynes et al., 2000). Ralph (2000) reported reductions in effective and maximum quantum yield of photosystem II in the aquatic grass species *Halophila ovalis* with exposure to atrazine, simazine, and diuron, while exposure to glyphosate had no effects. In concentrations of 0.2 mg/l of paraquat and 2 mg/l of glyphosate, Wong (2000) observed a significant reduction of the photosynthetic rate of algae measured by a differential respirometer. It appears that few studies have utilized fluorescence measurement for detection of 2,4-D injury in higher plants. However, it was reported that photosynthetic rates increased in algae exposed to low concentrations of 2,4-D, while exposure to levels of 2 mg/l and higher significantly decreased the photosynthetic rate (Wong, 2000). The use of chlorophyll *a* fluorescence to detect 2,4-D drift injury in cotton is unexplored.

Relatively recent advancements in chlorophyll fluorometry allow for a kinetic description of the fluorescence trace from ground state fluorescence (called the O step) to maximal fluorescence (the P step). J and I are intermediate steps in the fluorescence rise from ground state to maximal fluorescence. The use of OJIP, or chlorophyll fluorescence transient measurements, has become a widely used method of quantifying photosynthetic properties, as a large number of samples can be measured in a relatively short amount of time (Appenroth et al., 2001; Strasser et al., 2004; Strauss et al., 2006; Oukarroum et al., 2007; Yusuf et al., 2010; Stirbet and Govindjee, 2011). This analysis yields a wide array of photosynthetic based parameters and has been described as a way to measure structural or functional changes in photosynthetic performance (Stirbet and Govindjee, 2011), or plant vitality (Strasser et al., 2004; Oukarroum et al., 2007) in response to various factors (Srivastava et al., 1997; Srivastava et al., 1998; Srivastava et al., 1999; Strasser et al., 2004; Strauss et al., 2006). Readers are referred to Stirbet and Govindjee (2011), Strasser et al. (2004), and Strasser et al. (2000) for a more detailed overview of the parameters measured by OJIP analysis. The current study will focus on F_v/F_m, ϕ_{EO}, and PI_{abs}, which are calculated from the OJIP transient. Perhaps the most important parameter, in terms of utilizing a parameter for injury ratings, is PI_{abs}. This is the photosynthetic performance index and it reflects the conservation of energy from photons absorbed in photosystem II to be used for electron transport (Strasser et al., 2004; Strauss et al., 2006; Oukarroum et al., 2007; Stirbet and Govindjee, 2011). This index has been successfully applied in previous research as a screening tool for dark chilling tolerance in soybean cultivars (Strauss et al., 2006) and has been correlated with biomass production in beech trees exposed to different levels of ozone pollution (Clark et al., 2000).

Irrigation Management of Cotton in Conservation Tillage Utilizing a High Biomass Rye Cover Crop. Cotton is an important component to Georgia’s economy as the state ranked second in the nation with 2.47 million bales produced in 2011 at a value of 1.18 billion U.S. dollars (USDA – NASS, 2012a). Cotton was harvested on over 500,000 hectares during 2012 alone in Georgia (USDA – NASS, 2012b) and is the most widely grown field crop in the state. Water availability for agricultural purposes continues to be a source of concern for producers across the southeast, and Georgia in particular, as the state passed the Water Stewardship Act in 2010 increasing water use restrictions statewide (GDNR – EPD, 2011). Further action has been taken as the Georgia Environmental Protection Division suspended it’s consideration of some new farm water permits in several of the major cotton-producing counties in the state (GDA, 2012). With new issues regarding agricultural water usage arising, the competition over water rights and usage will continue to grow (Ward and Michelsen, 2002). As of the last agricultural census in 2007, there were over 121,000 hectares of irrigated cotton in Georgia (USDA – NASS, 2007). Given the variable climate and rainfall patterns observed during the growing season for cotton, which is typically May through September, irrigation and adequate water supply is critical for growers as more than half of the cotton acreage in Georgia will likely be irrigated by 2020 (Cai et al., 2010). Due to the arising challenges and expected increase in irrigated cotton acreage, it is critical that growers employ the most efficient irrigation and management practices for their operations.

Water is widely known to be one of the most limiting factors to agricultural production. Similar to most other agronomic crops, water stress has a detrimental effect on the physiological processes, growth, and development of cotton. Turner et al. (1986) observed decreases in leaf water potential, leaf conductance, and photosynthetic rates in cotton plants under water stress compared to plants that were fully irrigated. An optimal leaf area index throughout crop canopy development is beneficial to the growth and development of crops through maximizing light interception and acting as a photosynthetic source for fruiting sinks (Singh et al., 2006). Under water stress, the leaf area index of cotton is reduced compared to non-stressed plants (Turner et al., 1986; Orgaz et al., 1992) while water stress occurring late in the growing season increases the rate of decline of leaf area index, as well as total above ground dry matter (Gerik et al., 1996). Water stress has also been shown to have an impact on the morphology and phenological characteristics of cotton. In a study by Pace et al. (1999) that induced a 13 day drought period on cotton plants, plant height, leaf area, and dry weights of stems and leaves were reduced compared to control plants immediately following the drought period. Even after watering of the plants under the drought treatment was resumed, the plants continued to exhibit reductions in the aforementioned parameters for the remainder of the season (Pace et al., 1999), suggesting that recovery from periods of drought stress may not be achieved in some situations, illustrating the lasting negative impacts of a relatively short drought period on cotton. Pettigrew (2004a) found that irrigation increased the plant height, number of main stem nodes, and vegetative branches compared to cotton grown without irrigation. As the number of fruiting positions developed by the plant is directly related to the rate and amount of vegetative growth, this reduction in vegetative growth would most likely result in a yield reduction (Jordan, 1986).

Several studies have been conducted which documented negative effects on the yield components and lint yield of cotton under water stress. Water stress has been found to reduce the number of fruiting sights (Turner et al., 1986), the number of blooms (Guinn and Mauney, 1984a; Pettigrew, 2004a), and the number of bolls per plant in dryland or deficit irrigated conditions compared to fully irrigated cotton (Guinn and Mauney, 1984b; Morrow and Krieg, 1990). Gerik et al. (1996) determined that water stress reduced boll weight and the number of bolls per leaf area, while in two out of four years Pettigrew (2004a) observed a decrease in the number of bolls per unit ground area in water stressed cotton. Compared to non-irrigated plants, irrigated cotton tends to have a greater horizontal distribution of bolls, or bolls set on the second and third fruiting positions while non-irrigated cotton sets a greater percentage of bolls at the first position (Pettigrew, 2004a). Previous research that evaluated deficit irrigation or simulated episodic drought has shown that lint yield is reduced under these conditions compared to cotton that is fully irrigated (Turner et al., 1986; Gerik et al., 1996; Pringle and Martin, 2003; Pettigrew, 2004a; Whitaker et al., 2008; Gwathmey et al., 2011; Wen et al., 2013). It has also been observed that water deficits have a negative impact on fiber quality (Grimes and Yamada, 1982; Pettigrew, 2004a; Whitaker et al., 2008; Wen et al., 2013).

The magnitude of the negative impact water stress has on cotton yield is greatly influenced by the timing of the water stress in regard to the growth stage of the plant. Water stress occurring during the flowering and fruiting period will result in the most severe reduction in yield (Morrow and Krieg, 1990). This is widely regarded as the most sensitive growth stage of cotton, as both water and nutrients are at peak demand (Collins and Hake, 2012). Water stress during this period will reduce plant growth and the number of fruiting sites developed as well as increase boll shedding (Bauer et al., 2012b). Bednarz et al. (2003) determined that full irrigation during the flowering period increased yields by over 390 kg ha^{-1} compared to no irrigation.

Another characteristic of cotton growth and development that is influenced by water availability or water stress is maturity. As reproductive growth persists, the rate of development of main stem nodes slows as the upward progression of flowering approaches the terminal, which eventually ceases when the number of main stem nodes above a white flower (on the first position of a fruiting branch) reaches five, a stage known as physiological cutout (Oosterhuis et al., 1992; Bourland et al., 2001). Any bolls developed beyond this point generally contribute very little to the final yield (Oosterhuis et al., 1992). This response leads to a reduction in nodes above white flower (NAWF) when comparing cotton under reduced or no irrigation to fully irrigated plants, and results in accelerated maturity (Pettigrew, 2004a; Whitaker et al., 2008). The distribution of bolls on nodes is also affected by water stress as a greater percentage of bolls will be set on lower nodes in plants experiencing some sort of deficit or stress while there is a greater vertical distribution of bolls on higher nodes on well watered plants (Guinn and Mauney, 1984b; Gerik et al., 1996; Pettigrew, 2004a; Ritchie et al., 2009). Another measure of maturity that is accelerated when cotton is grown under deficit irrigation or water stress is time required to reach the number of nodes above cracked boll less than or equal to four ($NACB\leq4$) at which point harvest aids are generally applied (Gwathmey et al., 2011). This accelerated maturity in response to water stress potentially reduces the yield potential of cotton.

Although all cultivars of upland cotton are indeterminate in terms of growth habit (Ray and Richmond, 1966) it is generally accepted that there are two characterization of maturity available for commercial production. These are termed determinate and indeterminate, though may be referred to as short and long season, or early and late maturity, which indicates differences in the fruiting habit (Husman et al., 1996). Early season cultivars characteristically develop bolls over a shorter period of time compared to late season cultivars, and the majority of lint yield in early season cultivars is located at lower main stem nodes compared to late season cultivars, which have a more uniform vertical distribution of bolls that contribute to lint yield (Husman et al., 1996; Bednarz and Nichols, 2005). Early season cultivars typically perform better under conditions of optimal water availability and limited heat units or degree days (Rosenow et al., 1983; Snowden et al., 2013). However, due to a more abbreviated period of boll development, water stress will limit production and could result in significant fruit shedding from early season cultivars while full season cultivars would outperform early cultivars under these conditions (Rosenow et al., 1983; Husman et al., 1996; Snowden et al., 2013;).

While rainfall events in the Southeast are common during the cotton growing season, the amount and frequency of precipitation events varies year to year and have proven to be unpredictable and inconsistent, as optimal rainfall does not necessarily coincide with periods of critical water demand for each field. Additionally, episodic drought is often observed in many growing seasons in the southeastern U.S. This has been particularly true for Georgia which experienced a severe drought for several years during the late 1990s and early 2000s that causes diminished flow or even complete drying up of some streams in the state (Churchel and Batzer, 2006). For many cotton producing areas, including the Southeastern United States, irrigation is essential for consistently achieving acceptable yields and avoiding detrimental effects of water stress.

There are many tools, techniques, and methods available for scheduling irrigation so that the correct amount of water is applied when needed. Many of these rely on sensor based measurements to determine soil or crop water status for irrigation scheduling. It has been reported that while plant based sensors provide a more direct measurement of crop water status compared to soil based measures, the utilization of plant based measures may not yet be widely adopted (Jones, 2004). Alchanatis et al. (2010) found a relationship between thermal images of the crop canopy and leaf water potential values in cotton, however this technique has not yet been adopted by growers and consultants because of the high cost of the equipment required and the technical aspect of these measurements. There are also a variety of soil moisture sensors available to aid in irrigation scheduling, however the best-quality sensors or those that are typically used in research settings are again not practical for grower utilization due to the technical aspect and high cost of equipment (Vellidis et al., 2008). The software program, Irrigator Pro™, which was developed in collaboration between the USDA and the University of Georgia, is another irrigation scheduling tool available for growers (Nuti et al., 2009). The program uses inputs such as soil type, daily rainfall, crop growth stage, temperature, irrigation capacity, and crop variety to provide irrigation scheduling recommendations (Davidson, 1998; Morrison, 2005; Nuti et al., 2009).

Another method for irrigation scheduling is replacing evapotranspiration (ET), or the loss of water from the crop as transpiration and soil surface as evaporation combined (Allen et al., 1998). The foundation of this method is the calculation of the amount of evaporation from a crop, based on crop and soil characteristics, (ET_c) from the reference ET (based on environmental conditions) by the FAO Penman-Monteith equation and a crop coefficient (dependent on crop type, climate, and growth stage) (Allen et al., 1998).

A more common and relatively simpler method of irrigation scheduling that is used across a wide range of crops is the checkbook or water balance method. In this method, the water status of a field is maintained as an account, where rain or irrigation events are credited to the water balance and crop water use is considered withdraws from the water balance (Sassenrath and Schmidt, 2012). For Georgia grown cotton, crop water requirements by growth stage are made available through the cotton production guide (Table 1).

Coupled with efficient irrigation scheduling, another critical factor to cotton production in both irrigated and dryland situations, is maximizing the retention of applied or intercepted rainfall in order to provide the most benefit to the crop. Peters and Russell (1959) estimated that water loss due to evaporation from the soil surface in a row crop setting ranged between 50 to 70 percent of the water applied. The rate of evaporative loss of water from the soil surface is a factor of the evaporative demand as well as the amount of shading provided by the crop canopy (Al-Khafaf et al., 1978).

Limiting the amount of bare soil surface exposed to direct sunlight could increase both the amount of water, and the length of time that it will be available for uptake by the crop. Conservation tillage has become a popular method to reduce surface evaporation, and is characterized by the Conservation Tillage Information Center (CTIC) as a tillage and planting management system that contains crop residue covering 30% of the soil surface after planting (CTIC, 2002). It was reported in a 2005 survey that 53% of Georgia cotton growers used conservation tillage practices in their production system (Shurley, 2006). Numerous studies have found that soil moisture or water content is increased under conservation tillage (Blevins et al., 1971; Gantzer and Blake, 1978; Dao, 1993; Daniel et al., 1999b). It has also been reported that water infiltration rates increase in conditions where primary crops are grown with cover crops utilizing conservation tillage compared to conventional tillage (Dao, 1993; Bruce et al., 1995; Raper et al., 2000). Lascano et al. (1994) found that the soil moisture in strip tilled cotton planted into a cover of wheat stubble was significantly higher for five days after an irrigation application than the soil moisture in conventionally tilled cotton, while Mills et al. (1988) reported greater rainfall retention with conservation tillage. Landale et al. (1990) found the greatest increase in soybean yields in conservation tillage compared to conventional tillage under drought conditions. Increased infiltration and retention rates can reduce the amount of irrigation applications needed for cotton grown under irrigation, and illustrate the benefit to crops grown in dryland conditions where rainfall events are unpredictable and short term episodic drought conditions can develop.

Several different species of cover crops have been utilized in previous studies to determine the benefits they provide in a conservation tillage system. Ideally, a cover crop would produce high aboveground biomass so that soil surface coverage is maximized, water loss is minimal, and soil moisture and infiltration are optimal. Cover crop biomass yields vary, with the biomass production attributed to climate conditions. In the Southeast alone, results from multiple studies report biomass totals of several different crops ranging from 300 – 8900 kg/ha (Bauer and Busscher, 1996; Daniel et al., 1999a; Sainju et al., 2005; Schomberg et al., 2006; Balkcom et al., 2007; Price et al., 2009; Aulakh et al., 2012a; Bauer et al., 2012a). The studies above quantified the biomass generated for multiple common cover crops such as hairy vetch (Bauer and Busscher, 1996; Daniel et al., 1999a; Sainju et al., 2005; Schomberg et al., 2006;), black oat (Schomberg et al., 2006; Price et al., 2009), wheat (Daniel 1999a; Price et al., 2009), clover (Bauer and Busscher, 1996; Daniel et al., 1999a; Schomberg et al., 2006; Aulakh et al., 2012a;), lupin (Daniel et al., 1999a), winter pea, and oilseed radish (Schomberg et al., 2006). Additionally, all of the studies included rye, which has become a popular cover crop in the Southeast for several reasons. Rye biomass from the studies above ranged from 198 – to 7993 kg/ha. In the majority of studies comparing rye to other species of cover crop, either rye alone or a seed mixture of rye with another species produced the most biomass (Bauer and Busscher, 1996; Daniel et al., 1999a; Sainju et al., 2005; Schomberg et al., 2006; Aulakh et al., 2012a). The average biomass of rye that was documented in the aforementioned experiments excluding ones observing soil fertility treatments, was 3,477 kg/ha (Bauer and Busscher, 1996; Sainju et al., 2005; Schomberg et al., 2006; Balkcom et al., 2007; Price et al., 2009). Beyond the potential to produce high biomass, another advantage that rye has for a cropping system is its tolerance to cold, unlike other cover crop options such as black oat, oat, and wheat (Bauer and Reeves, 1999; Reeves et al., 2005).

Several beneficial effects have been reported from cotton grown with cover crops in a conservation tillage system. Taller plants (Bauer and Busscher, 1996; Bauer et al., 2010;), greater height to node ratios (Bauer et al. 2010) and a greater number of nodes (Wiatrak et al., 2005) have been observed in cotton grown in conservation tillage compared to conventional tillage systems. Reductions in root-knot nematodes (Bauer et al., 2010) and thrips (Manley et al., 2003; Olson et al., 2006;) have also been observed under conservation tillage, with greater thrips reductions found with higher biomass residue cover crops (Olson et al., 2006). Previous studies have also found that cover crop residues do not inhibit the cotton plant population (Schomberg et al., 2006; Wiatrak et al., 2006). Multiple studies have reported a yield increase in cotton planted into cover crop residues with conservation tillage compared to conventional systems (Bordovsky et al., 1994; Bauer and Busscher, 1996; Raper et al., 2000; Wiatrak et al., 2006; Bauer et al., 2010). It has also been observed that cotton planted into a rye cover yielded higher than cotton planted into other cover crop species (Bauer and Busscher, 1996; Schomberg et al., 2006).

Weeds are a constant problem for growers across all agricultural systems and require season-long management. This is particularly true for growers across the southeast where glyphosate resistant Palmer amaranth was first observed in 2005 in Georgia (Culpepper et al., 2006; Heap, 2015) and North Carolina (Heap, 2015). Glyphosate resistant crops have been widely adopted by growers since their release (Culpepper et al., 2008) due to the ability to easily achieve optimal weed control as traditional herbicide programs with fewer applications (Culpepper and York, 1998). Since the first discovery of resistance, glyphosate resistant Palmer amaranth has spread to over 2 million hectares (Sosnoskie and Culpepper, 2012) and is currently found in 25 states (Heap, 2015). The rapid spread of resistant Palmer amaranth is in part due to its ability to produce an average of 400,000 seeds per plant (Culpepper et al., 2007). Presence of Palmer amaranth in cotton can result in severe negative impacts to the crop. Morgan et al. (2001) observed a decrease in the canopy volume and biomass of cotton grown where Palmer amaranth was present. The existence of one Palmer amaranth per 9.1 to 10 meters of row was found to have the ability to reduce cotton yield 11 to 13 percent with higher weed densities of 8 to 10 per 9.1 to 10 meters of row reducing yield 47 to 92 percent (Rowland et al., 1999; Morgan et al., 2001), while Smith et al. (2000) found yield reductions ranging from 90 to 120 kg ha^{-1} in cotton plots with high densities of Palmer amaranth. Mechanical harvesting of cotton is also impacted by Palmer amaranth as harvest time is increased due to the removal of the weed from the harvesters and harvesting efficiency is reduced in cotton infested with Palmer amaranth resulting in a yield loss compared to weed free areas (Smith et al., 2000). Morgan et al. (2001)

observed that at higher densities of Palmer amaranth, there was the potential for damage to harvesting equipment, making mechanical harvesting unfeasible.

Cover crops have the ability to provide physical weed control by altering the environment around weed seed through blocking the interception of solar radiation and in some cases can provide chemical control through allelopathy (Creamer et al., 1996). Rye in particular is known to have allelopathic properties (Barnes and Putnam, 1986) and Ercoli et al. (2007) found that these properties are effective in the inhibition of germination of several different weed species. However, Sosnoskie et al. (2012) determined that it is unlikely that conservation tillage systems in Georgia would benefit from the allelopathic properties of rye as young fresh rye tissues at high concentrations are needed to achieve these benefits, and it would be unreasonable for individual growers to go through the process of obtaining the tissues and preparing them in the necessary fashion to utilize them in the manner required for allelopathic control of weeds. The typical management practices of conservation tillage in the Southeast are to kill the cover crop with herbicides and roll the residues prior to planting the primary crop. In this system the cover crop provides physical weed control, primarily through the obstruction of solar radiation required for the germination, growth, and development of weed seeds (Gallagher and Cardina, 1998a, 1998b; Cristaudo et al., 2007; Jha et al., 2010).

The capacity of cover crops to provide weed suppression has been illustrated by multiple studies, with rye being specifically successful (Putnam and DeFrank, 1983; Liebl et al., 1992; Zasada et al., 1997; Williams et al., 1998; Mafakheri et al., 2010;). Rye has been successful for control of Palmer amaranth (Culpepper et al., 2010; Sosnoskie and Culpepper, 2011; Aulakh et al., 2012a), more so than other cover crops (Norsworthy et al., 2011). Coupling a rye cover crop with other methods of weed control such as deep tillage (Sosnoskie and Culpepper, 2011;

Culpepper et al., 2010), herbicide programs (Reeves et al., 2005; Norsworthy et al., 2011), or a combination of tillage and herbicides (Aulakh et al., 2012a) has resulted in optimal weed control in cotton. It has been determined that employing a cover crop may allow for fewer herbicide applications while still providing compatible weed control to that found in a high input herbicide program (Reeves et al., 2005). It has been observed that greater weed control is accomplished with cover crops that produce a high amount of biomass, thus increasing the amount of residue present when the primary crop is planted (Norsworthy et al., 2011; Aulakh et al., 2012b). Previous studies have shown that rye is one of the best choices for a cover crop which will have the potential to produce substantial biomass for weed suppression (Williams et al., 1998; Culpepper et al., 2010; Mafakheri et al., 2010).

Physiological Impacts of Cotton Grown under Differing Irrigation and Tillage Practices. Water is widely regarded as one of the most limiting factors in crop production. Because of this, the response of plants to water stress has been a conventional research subject since the 1960's. In particular, the response of physiological processes in the plant to water deficit stress has been studied extensively. Essentially, water deficits in plants occur when the water lost from the leaf exceeds the uptake of water through the roots (Bray, 1997; Lawlor and Tezara, 2009). When exposed to water stress, the growth and development of plants is negatively affected (Mittler, 2006) however, several individual physiological responses occur that lead to this reduction in growth. The inhibition of growth is one of the first and most sensitive responses to water-deficit stress (Hsaio and Acevedo, 1974; Chaves et al., 2003). Growth inhibition occurs through a reduction in both cell expansion and cell division (Hsiao, 1973). Under conditions of water stress, pressure potential (Ψp) in plant cells is reduced, leading to a reduction in turgor and the ability of cells to expand (Bray, 1997). Stomatal closure is another response that plants exhibit early in response to water stress (Chaves et al., 2003). The closure of stomates in response to water stress is rapid so that water loss from the leaf is minimized (Lawlor and Tezara, 2009). Stomatal closure results in a reduction of transpiration and an increase in canopy temperature, the severity of which is determined by environmental factors (Hsiao, 1973). In turn, the closure of stomata alters several other physiological processes. When stomata close in response to drought stress, CO_2 diffusion to the carboxylation site is inhibited, thereby decreasing photosynthetic rates (Vaadia et al., 1961; Hsiao and Acevedo, 1974; Flexas et al., 2004; Saibo et al., 2008; Lawlor and Tezara, 2009). Other components of photosynthesis, such as the efficiency of photosystem II (Fv/Fm), electron transport rate (ETR), and the content of RuBP are negatively affected by water deficit (Saibo, 2008). However, the degree to which ETR is affected is dependent on the severity of the water stress the plant is experiencing (Medrano et al., 2002). It has been determined that stomatal conductance measurements show higher correlation with ETR than either relative water content or leaf water potential (Medrano et al., 2002). The water status of the plant is also affected by drought stress. The relative water content (RWC) of the leaf and the leaf water potential (Ψ_l) are reduced in response to water stress (Vaadia et al., 1961; Lawlor and Tezara, 2009). However, in terms of defining the level of stress the plant experiences, Ψ_l tends to be a better indicator of the actual effect on the water status of the plant, as RWC is less sensitive to mild and moderate water deficit stress (Hsiao, 1973). Additionally, for crops grown in summer months in climates known for high temperatures during the growing season, such as cotton in Georgia, water stress is often coupled with heat stress to further hinder plant growth and performance (Mittler, 2006).

Water stress can have a detrimental effect on several physiological processes in cotton. Previous studies have demonstrated decreased photosynthetic rates in water-stressed cotton compared to the rates of well-watered plants (Ackerson and Hebert, 1981; Turner et al., 1986; Ratnayaka et al., 2003; Carmo-Silva et al., 2012). Water stress has been shown to reduce the maximum efficiency of photosystem II (Fv/Fm) (Carmo-Silva et al., 2012; Ratnayaka et al., 2013) as well as the actual efficiency of photosystem II ($Ø_{PSII}$) (Carmo-Silva et al., 2012). However, Massacci et al. (2008) observed higher Fv/Fm and $Ø_{PSII}$ values in drought stressed leaves under low light intensity, while observing no difference in Fv/Fm and $Ø_{PSII}$ at high light intensity. A reduction in electron transport activity (Massacci et al., 2008; Carmo-Silva et al., 2012) as well as an increase in photorespiration (Massacci et al., 2008) have been observed when comparing water stressed and well-watered cotton leaves. However, other studies have reported higher electron transport rates in drought stressed cotton compared to fully irrigated cotton (Kitao and Lei, 2007; Zhang et al., 2011), as well as quantum yield of photosystem II in droughted plants compared to well-watered (Kitao and Lei, 2007). Pettigrew (2004b) reported higher electron transport rates and quantum efficiency of photosystem II in dryland cotton during morning measurements, while irrigated cotton had higher values in these measurements taken in the afternoon. Decreased stomatal conductance is another physiological response that has been observed in cotton under water stress, and has a direct effect on photosynthetic rate by reducing the amount of CO_2 entering the leaf. Stomates typically respond rapidly to water stress through reductions in stomata aperture to minimize the loss of water content from the leaf (Lawlor and Tezara, 2009). Measurements of stomatal conductance rates have been found to be physiological process most directly related to reductions in photosynthetic rates in response to mild to moderate water stress (Flexas et al., 2002; Medrano et al., 2002). The rate of decline in stomatal conductance may vary between cultivars under water stress (Carmo-Silva et al., 2012). Typically a decline in stomatal conductance is associated with a decline in leaf water potential in cotton when the plant is exposed to water stress (Radin, 1984; Turner et al., 1986). However, under prolonged water stress as mentioned above, it has been observed that stomata adapt to stay open under lower leaf water potentials than early in the stress period (Ackerson, 1980; Ackerson and Hebert, 1981). Leaf water potential impacts photosynthesis (Ackerson et al., 1977b) and conductance (Radin, 1984; Turner et al., 1986) in plants, and can be indicators of plant water status. Leaf water potential is widely accepted and utilized as a measure of water stress in crops. When compared to well-watered cotton, leaf water potential declines in cotton exposed to drought stress conditions (Ackerson et al., 1977a, 1977b; Ackerson and Hebert, 1981; Radin, 1984; Turner et al., 1986).

Pace et al. (1999) observed a recovery response in cotton in a study utilizing a stress and recovery period. After a ten day recovery period following 13 days of drought, the leaf area of plants in the drought treatment increased compared to measurements taken at the end of the drought period. This was attributed to the plant facilitating leaf area expansion through preferential photosynthate partitioning (Pace et al., 1999). An optimal leaf area index throughout crop canopy development is beneficial to the growth and development of crops through maximizing light interception and acting as a photosynthetic source for fruiting sinks (Singh et al., 2006). Other studies have observed a reduction in leaf area index (LAI) (Turner et al., 1986; Orgaz et al., 1992;), and specific leaf area (Carmo-Silva et al., 2012) due to water stress, while water stress occurring late in the growing season increases the rate of decline of leaf area index and reduces total above ground dry matter (Gerik et al., 1996). Pace et al. (1999) also reported shorter plant height, fewer nodes, and lower dry weight of leaves and stems at the end of the recovery period. This suggests that the ability of cotton to recover from periods of drought stress may not be achieved in some situations, and illustrates the long term effects that a mild to moderate drought period can have on the growth and development of cotton.

The ability of cotton to acclimate, or develop a tolerance to water stress over time has also been observed in previous studies. Ackerson and Hebert (1981) subjected cotton plants to consecutive cycles of water stress and recovery, and while initial measurements showed reductions in photosynthetic rates, the plants adapted and at the last two treatment cycles, the stressed plants had higher photosynthetic rates than the controls. This was the result of the stomata in the stressed plants remaining open at lower leaf water potentials in drought stressed plants, allowing for continued carbon assimilation (Ackerson and Hebert, 1981). This was accomplished through osmotic adjustment in the stressed plants, which resulted in the leaves maintaining turgor at lower leaf water potentials than the leaves in the control (Ackerson and Hebert, 1981).

Along with these crucial physiological processes, water stress in cotton has also been found to decrease nitrate reductase activity in leaves (Ackerson et al., 1977b) and increase canopy temperatures (Carmo-Silva et al., 2012) which is typically linked with stomatal closure. Physiological response to drought may vary by cultivar, as some cultivars are more drought-sensitive or tolerant than others (Carmo-Silva et al., 2012).

Another method researchers have used to study and define the effects of water stress on plants is diurnal measurements, or several measurements at intervals over an entire day. Past research has documented decreases in conductance levels as early as two hours after sunrise under drought conditions (Jordan and Ritchie, 1971). This reflects the findings mentioned previously that stomatal conductance is one of the most sensitive physiological processes in response to water stress. In both well-watered plants and those under drought stress, higher stomatal conductance rates have been observed during the morning, with rates declining in the afternoon as radiation and ambient temperatures increase (Massacci et al., 2008). Morning conductance rates in drought-stressed plants are typically lower, and the afternoon decline is slower, compared to well-watered plants, due to the lower amount of water available to the plants undergoing stress (Massacci et al., 2008). However, Ackerson (1981) observed higher conductance rates in the morning and early afternoon, with a steeper decline in rates during the late afternoon, in drought-stressed compared to well-watered mainstem leaves on the fifth node in cotton. For leaves on the eighth node, the morning levels were similar, with the drought-stressed leaves maintaining higher conductance rates for a longer period of time compared to well-watered leaves during the afternoon decline in conductance rates (Ackerson, 1981). These findings were attributed to the ability of cotton to acclimate to water stress (Ackerson, 1981; Ackerson and Hebert, 1981). The photosynthetic rates in the morning did not follow the same trend, as the drought-stressed leaves exhibited lower morning photosynthetic rates than the well-watered plants in the morning, while the rates of photosynthesis in the afternoon were higher and maintained for a longer period of time in leaves at both the fifth and eighth node in the droughted leaves compared to the well-watered leaves (Ackerson, 1981). This also illustrates the importance of adequate water availability, as even with the ability of the stomata to remain open under water deficit conditions, the photosynthetic function of the plant is still impaired.

Water deficit has been observed to impact the diurnal behavior of leaf water potential (Jordan, 1970). A typical diurnal leaf water potential pattern for cotton is for it to peak overnight at approximately midnight and steadily decline as the sun rises with the lowest point occurring in the early afternoon, followed by a steady rise as the sun sets in the evening (Jordan and Ritchie, 1971). Compared to well-watered cotton, it has been found that the leaf water potential is reduced in cotton under water stress throughout the diurnal measurement period (Ackerson et al., 1977a; Grimes and Yamada, 1982). This is significant as it illustrates that unlike the findings from previous studies showing the ability of cotton to acclimate to water stress after exposure to multiple drying cycles (Ackerson, 1981; Ackerson and Hebert, 1981), leaf water potential is significantly reduced in cotton when exposed to one period of water stress. This represents a situation that would likely be present under normal growing conditions, while continuous and uniform cycles of drought and recovery are not typical climate patterns, and are not typical of the irrigation schedules used by producers.

Research of diurnal responses to water deficits has been performed in other crops with similar results to those mentioned above for cotton. Pear (Klepper, 1968) and onion (Millar et al., 1971a) were reported to have a similar diurnal pattern of leaf water potential to that of cotton, with the lowest values reached at midday with an increase towards sunset. A reduction in leaf water potential and an increase in leaf temperature have been observed in barley grown in water deficient conditions compared to well-watered conditions (Millar et al., 1971b). Stomatal conductance is directly affected by leaf water potentials, as conductance rates decline with decreasing leaf water potential, emphasizing the role of plant water status on maintaining stomatal conductance and thus, photosynthesis, and other physiological functions in the plant (Millar et al., 1971a).

Recently, producers have adopted the use of conservation tillage practices, which includes employing a cover crop, typically a small grain or legume winter species grown in the winter and early spring months. In some cases, the cover crop is allowed to persist until up to a few weeks prior to planting the primary crop, and is killed with an herbicide and rolled to provide residue for soil coverage. For other winter cover crops, such as wheat, the cover crop is harvested prior to the planting of the primary crop and the remaining stubble provides a degree of soil coverage and residue for the following growing season. Multiple studies have determined that soil moisture or water content is increased under conservation tillage and with the utilization of a cover crop (Blevins et al., 1971; Gantzer and Blake, 1978; Dao, 1993; Daniel et al., 1999b). While there appears to be little previous research on the effect of a cover crop on physiological processes of the subsequent crop, there are a few select studies that have reported the response of some processes. During a dry period, Cox et al. (1990) found lower carbon exchange rates and higher stomatal resistance in corn grown in no tillage conditions compared to conventionally tilled corn, while the carbon exchange rates and stomatal resistance values were similar once the corn was returned to well-watered conditions. In a two year study, Singer et al. (2007) utilized tillage treatments by employing either tillage with a chisel plow, moldboard plow, or no-tillage and found no differences in carbon exchange rates and stomatal conductance measurements between the tillage treatments in either year. Al-Darby et al. (1987) found no difference in leaf water potentials between corn grown under a variety of tillage practices and no-tillage. However, the corn in this study received adequate rainfall throughout the growing season and it is likely that no stress existed to elicit a physiological response. It has also been reported that the leaf area index of corn does not show a response to tillage treatments (López and Arrúe, 1997). Delays in phenological development (Cox et al., 1990) and early season growth (López and Arrúe, 1997), as well as reductions in biomass (Cox et al., 1990) have been reported when comparing no-tillage to conventional tillage. It is important to note, however, that corn was the focus of all the aforementioned studies, and that they were performed in New York (Cox et al., 1990), Iowa (Singer et al., 2007), Wisconsin (Al-Darby et al., 1987), and Spain (López and Arrúe, 1997). There doesn't appear to be any work on the effect of a cover crop on the physiological response of cotton grown under water stressed conditions. In terms of phenological and morphological parameters, several studies have reported beneficial results of cover crops compared to conventionally-grown cotton (Bauer and Busscher, 1996; Wiatrak et al., 2005; Bauer et al., 2010). Furthermore, in regards to yield, previous studies have shown increases in cotton yield under conservation tillage compared to conventional tillage (Bordovsky et al., 1994; Bauer and Busscher, 1996; Raper et al., 2000; Wiatrak et al., 2006; Bauer et al., 2010). It is also important to note that several studies observed yield increases with cover crops in dryland conditions (Bordovsky et al., 1994; Bauer and Busscher, 1996; Raper et al., 2000; Bauer et al., 2010), indicating that the utilization of cover crops results in greater retention of rainfall in the soil for longer periods of time leading to increased yields compared to conventional tillage in dryland conditions.

Influence of Leaf Pubescence and Defoliation Strategy on Defoliation and Fiber Quality of Cotton. Timing of harvest is an important issue for producers of indeterminate crops such as cotton, as proper timing is critical to maximizing the yield and profitability of the crop. The development of bolls takes place over several weeks (Buxton et al., 1973), so a balance must be met between harvesting to maximize the retention and capture of older bolls that mature earlier in the season, and the later developing young bolls that are beginning to reach maturity at harvest. Improving the efficiency of harvest activities, as well as lint quality, through defoliation has been a goal of cotton producers and the focus of studies attempting to improve the efficiency of defoliation practices since the early 1950s (Brown, 1953). Because cotton is a perennial that is grown and managed as an annual for agronomic benefits, eliminating green, live plant material and the amount of dead plant material that contaminates seed cotton at harvest is a major issue for obtaining optimal harvest efficiency and lint quality (Colwick et al., 1984). When performed properly, defoliation will reduce the amount of leaf and other plant material (referred to as trash) in the harvested seed cotton, further reducing damage to fiber in the ginning process by lessening the amount of cleaning operations required for cleaner lint (Brecke et al., 2001), as well as allowing for earlier harvest that increases the ease of mechanical harvesting operations while decreasing yield losses due to weathering (Siebert et al., 2006). The rate of dehiscence of bolls developed late in the season is also increased through the use of harvest aids, permitting an earlier harvest (Cathey et al., 1982). While many factors influence the effectiveness of cotton defoliation, including environmental, plant growth characteristics, and the types of products used, this review will focus on the effect of leaf pubescence and defoliation practices on defoliation success and trash grade.

Leaf hairs, or thrichomes, are evolved traits developed by plants that serve as a defense mechanism against damage due to herbivore feeding from insects (Butler et al., 1991; Huttunen et al., 2010). However, in terms of the effect on cotton production and ginning practices, leaf pubescence can have a deleterious effect in regard to defoliation success and lint quality due to trash content because of the pubescent plant material becoming entangled in the lint. Novick et al. (1991) found less total trash and a greater ability to clean leaf trash out of lint in semi-smooth (lower levels of leaf pubescence) cultivars compared to those with greater leaf pubescence. The occurrence of motes, which are immature seed or underdeveloped fiber in the lint (Davidonis et al., 2000) was also reduced in semi-smooth cultivars (Novick et al., 1991). An increase in the amount of trash in ginned lint has been observed by multiple other previous studies, and is in general the major detriment to leaf pubescence in cotton (Ramey, 1962; Smith, 1964; Wanjura et al., 1976;). A study by Bechere et al. (2011) showed that for one cultivar, the required ginning energy was increased for a hairy leaf (high levels of leaf pubescence) type compared to a smooth leaf (very low levels of leaf pubescence) type. While the amount of trash in harvested seed cotton may not differ between smooth and hairy leaf cultivars, trash from smooth leaf cultivars is easier to remove in the ginning process and lint from smooth leaf cultivars has been reported to have a higher grade than lint from hairy leaf cultivars (Colwick et al., 1984). Due to these factors, leaf smoothness has been one goal of cotton breeding to improve lint quality (Colwick et al., 1984).

Defoliants used in cotton function by initiating a hormonal response in the base of the petiole, termed the abscission zone, resulting in secretion of chemicals that degrade cell walls which leads to the leaf falling off the plant (Cathey, 1986). The performance of defoliators is inhibited if applications are made at improper rates. If too much is applied, the hormone that is needed to degrade cell walls will not be released as the plant tissues that contain this hormone will be killed, while if too little is applied, the leaf will not fully abscise from the plant (Cathey 1986). Optimal performance of defoliants occurs when applications are made to plants that are mature with healthy leaves, with no excess water or nutrients, while avoiding water stress, and while warm temperatures are present (Brecke et al., 2001).

Another form of harvest aid used on cotton is herbicides, or other chemicals that act as desiccants, which can lead to the death of the entire plant and prevent the formation of an abscission layer (Bovey and Miller, 1968; Brecke et al., 2001). Desiccation of cotton results in what is termed “leaf stick” or the dead leaves remaining on the plant, and is also seen when high rates of defoliants are utilized (Stahler, 1953; McMeans et al., 1966; Brecke et al., 2001; Shaw, 2002;). The result of optimal defoliation is the removal of green leaf material from the plant prior to harvest, while ensuring that cell and tissue damage does not occur, as the plant must be alive for an abscission layer to form and defoliation to occur (Stahler, 1953; Cathey, 1986;

Stichler et al., 1995; Clark and Carpenter, 1996). Without the formation of an abscission layer, leaf stick will occur with desiccation and trash grades in desiccated cotton will increase (Shaw, 2002). Leaf death due to the application of desiccants has been observed to occur six to seven days after application (Bovey and Miller, 1968; Clark and Carpenter, 1996), while the optimal effectiveness of defoliants, measured by leaf drop, has been observed to occur 17 days after application (Clark and Carpenter, 1996).

Previous studies have observed an effect on yield from differing harvest aid application practices. Cathey et al. (1982) reported higher yields in cotton that was defoliated compared to desiccated plants, while Brecke et al. (2001) found that cotton bolls continued to increase in size and weight after application of a defoliant and predicted that the increase of these yield components would most likely not be found in desiccated cotton.

Objectives

Impact of Sub-Lethal Rates of 2,4-D at Various Growth Stages of Cotton. Through this study, we project that we can quantify the effect of sub-lethal 2,4-D drift and tank contamination rates on the growth, development, and yield of non-tolerant cotton, as well as determine growth stages that are the most vulnerable. Due to the different management schedules producers are on in terms of planting and treating cotton with herbicides, multiple pre-bloom, bloom, and peak bloom growth stages will be targeted for simulated drift applications. Visual injury ratings will be taken to determine the relationship between injury symptoms and yield loss. Plant mapping will also be performed to evaluate the effect of 2,4-D on reproductive development and crop maturity.

Potential for Utilizing Chlorophyll a Fluorescence to Detect Injury and Predict Yield Loss from Sub-Lethal Rates of 2,4-D on Cotton. From the physiology perspective of the 2,4-D study, our aim is to quantify the effect of sub-lethal rates of 2,4-D on the chlorophyll *a* fluorescence parameters Fv/Fm, ϕ_{EO}, and PI_{abs} of cotton at different growth stages. An additional objective of this study was also to determine if any of the responses of these physiological parameters could be correlated to observed yield reductions. We hypothesized that if any of these parameters could be identified as accurate predictors of yield loss, with the same or better accuracy compared to the visual estimates that have been used in the past, then the potential exists for the utilization of OJIP measurements to quantify injury and estimate yield loss of 2,4-D drift injury in cotton. If a high correlation exists between OJIP parameters and yield reductions, set points or thresholds could be instituted depending on the level of yield loss expected to occur at varying levels of these fluorescence values. Combining a set point as defined by fluorescence parameters with the ability to measure large numbers of plants in a relatively short amount of time, could allow for rapid determination of the intensity of injury from drift and a more accurate prediction of expected yield loss.

Irrigation Management of Cotton in Conservation Tillage Utilizing a High Biomass Rye Cover Crop. The conservation tillage cover crop study was evaluated from both an agronomic and physiological perspective. The agronomic objectives of this research were to document and quantify potential water savings or potential crop growth or yield improvements in cotton, under various irrigation schedules, resulting from a heavy rye residue tillage system.

Physiological Impacts of Cotton Grown under Differing Irrigation and Tillage Practices. From the physiological aspect of this study, the objective of this study was to compare and contrast the physiological responses of cotton to a high residue rye cover crop, compared to conventional tillage, in both irrigated and non-irrigated conditions. A variety of plant water status and physiological measurements will be performed to determine the response of cotton grown under the different management and irrigation systems. It is expected that the increased levels of plant available water that have been documented in past studies of cover crops may allow for more efficient use and storage of precipitation events and irrigation applications, resulting in a reduction in the number or duration of stress periods in cotton.

Influence of Leaf Pubescence and Defoliation Strategy on Defoliation and Fiber Quality of Cotton. The objectives for evaluating the of influence of leaf pubescence and defoliation strategy on fiber quality were to determine if higher leaf grades result primarily from leaf pubescence characteristics inherent to genetics of the cultivar or were primarily the product of aggressive defoliation practices. An additional objective was to determine if the influence of cultivar leaf pubescence and defoliation practices was additive or interactive. We hypothesized that the use of smooth leaf cultivars, defoliated at recommended application rates, will result in higher defoliation success and lower leaf trash grades in harvested lint, resulting in higher lint quality.

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CHAPTER 2

COTTON STAGE OF GROWTH DETERMINE SENSITIVITY TO 2,4-D[1]

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[1] S.A. Byrd, G.D. Collins, A.S. Culpepper, D.M. Dodds, K.L. Edmisten, D.L. Wright, G.D. Morgan, P.A. Baumann, P.A. Dotray, M.R. Manuchehri, A. S. Jones, T.L. Grey, T.M. Webster, J.W. Davis, J.R. Whitaker, P.M. Roberts, J.L. Snider, and W.M. Porter. To be submitted to *Weed Technology*.

Abstract

The impending release of Enlist^{TM} cotton and soybean cultivars will likely increase the use of 2,4-D which has raised concerns over potential off-target movement or tank contamination issues to susceptible cotton. An experiment was conducted at 12 locations across the cotton belt during 2013 and 2014 to determine the impact of a simulated 2,4-D drift application (2 g ae ha^{-1}) or tank contamination application (40 g ae ha^{-1}) would have on cotton during six different growth stages. Cotton growth stages included 4-leaf (4-lf), 9-leaf (9 lf), first bloom (FB), FB+2wk, FB+4wk, and FB+6wk. Visual epinasty from 2,4-D was far more pronounced with applications during vegetative growth stages; injury exceeded 50% at 7 of the 8 locations where visual injury was evaluated when either rate of 2,4-D was applied to 4- or 9-lf cotton. As cotton matured beyond the FB+2wk stage, visual injury from the drift application was less than 10% at 7 of 8 locations and for the contamination rate injury was less than 25% at 7 of 8 locations. The maximum level of epinasty from vegetatively treated plants occurred 4 to 5 wk after application while the mature cotton displayed maximum epinasty within a wk. Importantly, yield loss did not correlate with visual symptomology but rather more closely followed effects on cotton boll set. The contamination rate of 2,4-D applied to 9-lf, FB, or FB+2wk cotton had the greatest effect across locations reducing the number of bolls per plant when compared to the control. This was likely, in part, a response to boll injury noted with these treatments. The contamination rate of 2,4-D applied at FB+4 wk or later did not influence the number of bolls per plant and only at 4 locations did the application reduce the number of bolls per plant when applied at the 4-lf stage. A reduction of boll number per plant was not detectable with the drift rate of 2,4-D except at 4 locations when applied only at the FB stage of growth. Similar to boll set, yield was influenced by 2,4-D rate and stage of cotton growth. Loss in yield of greater than 20% occurred

at 5 of 12 locations when the drift rate was applied between 4-lf and FB+2wk (highest impact at FB). For the contamination rate, yield loss was observed at all 12 locations; averaged over these locations yield was 45, 58, 66, 45, 16, and 7% below that noted in the control when applied at 4-lf, 9-lf, FB, FB+2 wk, FB+4 wk, and FB+6 wk, respectively. Results suggest the greatest yield impact from 2,4-D would occur between 9-lf and FB+2wk and the level of impact will be influenced by 2,4-D rate and environmental conditions as they influence the progress toward boll development and plant recovery.

Introduction

Cotton tolerance to 2,4-D is conferred by the insertion of a gene that codes for the enzyme aryloxyalkanoate dioxygenase. Plants transformed to include this gene can metabolize 2,4-D to a non-lethal form (Richburg et al. 2012). This technology is being commercialized through the Enlist^{TM} line of cotton, soybean, and corn products from Dow AgroSciences (Dow AgroSciences 9330 Zionsville Road, Indianapolis, IN 46268). Commercialization of this technology in soybeans has occurred in some U.S. states with further adoption for soybeans and cotton expected during 2016.

Discovered in secret during World War II as a potential chemical weapon, 2,4-dichlorophenoxyacetic acid was the first selective herbicide widely used in agriculture (Peterson 1967). Since that time researchers have demonstrated control of a vast array of weed species, including Palmer amaranth (*Amaranthus palmeri*), horseweed (*Conyza canadensis*), sicklepod (*Senna obtusifolia*), giant ragweed (*Ambrosia trifida*), common waterhemp (*Amaranthus rudis*), common lambsquarters (*Chenopodium album*), velvetleaf (*Abutilon theophrasti*) , red morningglory (*Ipomoea coccinea*), hairy vetch (*Vicia villosa*), crimson clover (*Trifolium incarnatum*), cocklebur (*Xanthium commune*), Pennsylvania smartweed (*Polygonum* *pennsylvanicum*), and Jimsonweed (*Datura stramonium*) (Jha and Norsworthy, 2012; Kruger et al., 2010; Kruger et al., 2008; Lancaster et al., 2005; Robinson et al., 2012; Siebert et al., 2004; White and Worsham, 1990; Williams et al., 1960).

Adoption of 2,4-D in Enlist^{TM} technology in a given area will be influenced by yield potential of the crop, weed species infesting fields, and most notably the ability of growers to mitigate off-target movement of 2,4-D (Mortensen et al., 2012; Riar et al., 2013; Egan et al., 2014). Although Enlist^{TM} cotton is tolerant to 2,4-D (Johnson et al., 2012b; Dow AgroSciences 2015), all other cotton cultivars including cotton tolerant to dicamba are extremely sensitive to the herbicide with reports of cotton injury due to 2,4-D drift dating back to the 1940's (Staten, 1946). Multiple studies have reported that exposure to 2,4-D resulted in visual injuries in cotton; the severity of injury increased at earlier growth stages and at higher herbicide concentrations (Sciumbato et al., 2004; Marple et al., 2007; 2008; Everitt and Keeling, 2009; and Johnson et al., 2012a). Visual injury symptoms observed on cotton exposed to 2,4-D include leaf cupping, strapping, chlorosis, twisting of petioles, stem epinasty, stunting of growth, and loss of apical dominance (Marple et al., 2008; Everitt and Keeling, 2009; Johnson et al., 2012a). The primary tool utilized to quantify the magnitude of injury has been visual ratings of these symptoms following exposure (Sciumbato et al., 2004; Marple et al., 2008; Everitt and Keeling, 2009). Marple et al. (2007; 2008) also noted peak injury occurring at 28 days after application and that visual injury ratings taken during this time period were highly correlated with yield loss.

Most of the previous research utilized simulated drift treatments, or sublethal rates (0.28 – 280 g ae ha^{-1}), of 2,4-D to illustrate the effect of off-target movement of the herbicide on cotton at vegetative to early bloom growth stages (Marple et al., 2007; 2008; Everitt and Keeling, 2009; Johnson et al., 2012a). Due to the large planting window for cotton and the extremely large potential window for applications of 2,4-D in Enlist^{TM} technology, the objective of this experiment was to better understand how cotton growth stage influenced cotton response to 2,4-D when treated with a simulated drift rate or a simulated tank contamination rate.

Materials and Methods

Field experiments were conducted at 12 locations across the cotton belt during 2013 and 2014, with site specifics details provided in Table 2.1. The cultivar 'PhytoGen 499 WRF' (Dow AgroSciences, Indianapolis, IN) was planted across all locations and years following standard production practices for each location (Albers and Reinbott, 1994; Wright et al., 2014; Collins et al., 2015; Edmisten et al., 2015; Mississippi State University Extension Service, 2015a; Texas A&M AgriLife Extension, 2015). Plots were 12.2 meters long and consisted of four rows of cotton, with row spacing varying between 91 to 102 cm apart depending on location. Crop management and inputs followed the recommended practices for each location or region.

The factorial treatment arrangement included six cotton growth stages and two rates of 2,4-D (Weedar® 64, Nufarm Americas Inc. 4020 Aerial Center Parkway, Suite 101, Morrisville, NC 27560). Growth stages included four leaf (4-lf), nine leaf (9-lf), first bloom (FB), FB+2wk, FB+4wk, and FB+6wk. Rates of 2,4-D applied included a simulated drift rate at 2 g ae ha^{-1} and a simulated tank contamination rate at 40 g ae ha^{-1}; these rates were 1/421 and 1/21 of the recommended use rate at 841 g ae ha^{-1}, respectively . The two rates used in this study fell within the range of rates used by Johnson et al. (2012a), and were similar to the rates representing particle drift and misapplication in Egan et al. (2014). Applications of 2,4-D were made with a CO_2 backpack sprayer calibrated to deliver 140 L ha^{-1} at 180 kPa. Applications were made only to the center two rows of each plot, with the outer rows serving as a border between plots. A non-treated control void of 2,4-D was included for comparisons. The FB treatment was excluded from the 2013 New Deal, TX location.

Visual injury evaluations were taken throughout the season at the Moultrie and Tifton, GA, Lewiston, NC, and New Deal and Snook, TX locations using a scale of 0 (no injury) to 100% (complete crop death). Specific symptoms evaluated included leaf curling, petiole and stem epinasty, stunting, and chlorosis (Marple et al., 2008; Everitt and Keeling, 2009; Johnson et al., 2012a).

At crop maturity based on the non-treated control, cotton was defoliated following extension recommendations for each specific location (Albers et al., 1994; Leon et al., 2013; Kelly et al., 2014; Collins et al., 2015; Edmisten et al., 2015; Mississippi State University Extension Service, 2015b). After defoliation applications were made, and prior to harvest, 10 consecutive plants from each plot were removed by cutting the main stem directly above the soil surface and were utilized for plant mapping measurements. The plant was divided into 0.3 m sections of the main stem, and the number of open and closed bolls was quantified from each section. Bolls were attributed to the 0.3 m section in which the reproductive branch they were located on originated. Plant mapping data was collected at 11 locations with the one exception being Mississippi in 2013. Plots were harvested at the end of the season with a two row plot harvester and seed cotton yield was measured after harvest.

Treatment means for cotton yield at each location were expressed as a percentage of the non-treated control. Data were analyzed using PROC MIXED with the pdmix 800 macro included (Saxton, 1998) and treatment means were separated by Fisher's Protected LSD at an alpha level of ≤ 0.05 using SAS 9.4 software (SAS Institute Inc., Cary, NC). The main effect of location was significant when analyzed for the entire data set (data not shown). However, locations were able to be grouped into three categories because of similarities of cotton response to 2,4-D treatments. The three categories included Group I having the lowest level of yield loss, Group II having a moderate level of yield loss, and Group III having the greatest degree of yield loss. Locations included in group I were New Deal, TX (2013 and 2014), Starkville, MS 2014, and Snook, TX 2013. Group II consisted of Quincy, FL 2014, Snook, TX 2014, and Tifton, GA (2013 and 2014). Group III consisted of the Portageville, MO 2014, Lewiston, NC 2014, Starkville, MS 2013, and Moultrie, GA 2013.

Results and Discussion

Visual Injury Evaluations. Visual evaluations of crop injury are the most common method utilized to detect and quantify the severity of 2,4-D injury (Sciumbato et al., 2004; Marple et al., 2008; Everitt and Keeling, 2009). Exposure to 2,4-D produces obvious and distinct injury, particularly to actively growing vegetative portion of the plant, including leaf cupping and strapping and epinasty of petioles and stems (Marple et al., 2008; Everitt and Keeling, 2009; Johnson et al., 2012a). The highest level of 2,4-D injury observed across all locations occurred early in the season, during the pre-bloom growth stages (Table 2.2). Across all 3 location groups, injury from 4- and 9-lf applications ranged from 45 to 64% with the simulated drift rate and 58 to 83% with the simulated tank contamination rate. Maximum injury was observed 4 to 5 wk after application (data not shown). Less injury was observed within each grouping with applications made to cotton at FB and later as compared to vegetatively growing cotton (Table 2.2). Group I locations noted less than 12% injury with either rate of 2,4-D applied to cotton in the FB stage of growth compared to 21 to 45% with groups 2 and 3. Differing responses may have been a result of cotton in group I locations transitioning more aggressively into reproduction as compared to more vegetative growth following treatments. Applications made after FB noted injury ranging from 1 to 8% with the drift rate of 2,4-D across groups (Table 2.2). Differences across location groups were noted with the contamination rate of 2,4-D with 1 to 10% injury noted with group I, 9 to 24% injury with group II, and 18 to 25% injury with group III. Lower levels of injury with group I again suggest less vegetative growth was occurring at these locations. Maximum levels of injury, although low, were detected most often within a week of application (data not shown).

Plant Mapping. Previous studies have demonstrated that plant mapping provides valuable insight on vegetative and reproductive development of the cotton plant in response to stress (Constable, 1991; Plant and Kerby, 1995; Jones and Snipes, 1999; Pettigrew, 2004). Plant mapping was used to further describe the observed treatment differences due to application timing and rate of 2,4-D. There are two components that were observed to have a direct effect on cotton yield, the number of total bolls per plant (a measure of the effect of 2,4-D on the development of reproductive structures), and the percent of open bolls present at harvest (a measure of delayed maturity).

Although the number of bolls was counted from multiple 0.3 m regions of each plant (Fig. 2.1), few differences were noted within regions (data not shown). In contrast, viewing the total number of bolls per plant did note significant treatment effects (Table 2.3). The contamination rate of 2,4-D applied to cotton at 9-lf, FB, and FB+2wk had the greatest impact reducing the number of bolls per plant when compared to the control. This was likely, in part, a response to boll injury noted with these treatments (Fig. 2.3). The effect of 2,4-D on the development of bolls reflects previous work of Marple et al. (2007) who reported a reduction in flower development and increased flower abortion from 2,4-D exposure. The contamination rate of 2,4-D applied at FB+4wk or later did not influence the number of bolls per plant and only with Group I did this 2,4-D rate reduce the number of bolls per plant when applied at the 4-lf stage of growth. Impact of boll number per plant was not influenced by the drift rate of 2,4-D except only with Group III and only when applied at the FB stage of growth.

The percent of open bolls per plant was not influenced with applications made at FB or later within any group (Table 2.3). The greatest reduction in open boll percentages, thereby delaying crop maturity, was noted with 4- and 9-lf applications. The delayed maturity observed in the current study is similar to the findings of McIlrath et al. (1951) who reported a delay in boll development due to 2,4-D exposure when applied to cotton during the early bloom stage of growth. However, the total number of bolls is more indicative of yield than measurements of maturity such as open boll percentages, as Krieg (2000) reported that the number of bolls accounts for greater than 85% of the yield variability in cotton.

Yield. Yield loss was noted at all locations as influenced by rate and cotton stage of growth. Similar to injury, Group I had less overall yield loss while Group III had the most (Table 2.2). For Group I, the drift rate of 2,4-D did not influence yield when compared to the non-treated control. For the contamination rate, yield loss of 48 to 51% was noted with applications at 4 and 9-lf while 16 to 28% yield loss was noted with FB and FB+2 wk applications. Applications to more mature cotton did not influence yield within this group of locations. Similar to Group I, minimal instances of yield loss was noted from the drift rate of 2,4-D for Group II locations, with a 19% reduction occurring at FB applications being the only significant occurrence. However, the contamination rate applications reduced yields at all timings except the most mature one at FB+6 wk. Yield loss was greatest with applications at FB reducing yield 81%. Loss of 76%, 46%, 34%, and 19% occurred with contamination rate applications on cotton at FB+2 wk, 9-lf, 4-lf, and FB+4 wk, respectively. For Group III, both rates of 2,4-D reduced yield when applied from 4-lf through FB+2 wk; with the greatest level of impact occurring at FB. The contamination rate of 2,4-D also reduced yield when applied at FB+4 wk but to a less degree than earlier applications. Overall yield impact was greatest with Groups II and III with applications made near FB while Group I indicating greater sensitivity at 9- and 4-lf stages. This result along with injury and percent open boll percentages continue to suggest Group I included locations that progressed from vegetative to reproduce bolls the most rapidly.

When considering all locations, the contamination rate of 2,4-D reduced cotton yield 83, 92, 82, 83, 33, and 8% of the time when applied to 4-lf, 9-lf, FB, FB+2wk, FB+4wk, and FB+6wk, respectively. Yield loss from the drift rate of 2,4-D was observed 8, 25, 55, 25, 17, and 8% of the time with applications at 4-lf, 9-lf, FB, FB+2wk, FB+4wk, and FB+6wk, respectively. The amount of yield loss also indicated that the contamination rate had the greatest negative impact on yield when applied during FB, FB+2wk and 9-lf when compared to other application timings. For the drift rate, greatest impact occurred at FB followed by 9 lf and FB+2wk. Yield results suggest the greatest impact from 2,4-D would occur between 9-lf and FB+2wk and the level of impact will be influenced by 2,4-D rate and environmental conditions as they influence the progress toward boll development.

The level of visual cotton injury, plant maturity effect, boll abortion, and yield loss from potential 2,4-D drift, volatility and drift, or tank contaminations will be influenced by 2,4-D rate, cotton maturity, and environmental conditions that occur after the incident. Visual injury, primarily epinasty, is far more pronounced when cotton is damaged by 2,4-D in a vegetative stage of growth and lessens as the cotton becomes more mature. Often with visual epinasty, the peak level of injury for vegetatively growing plants does not appear until 4 to 5 wk after the incident. In contrast, more mature cotton containing a significant boll set will display minimal epinasty with maximum levels often observed by 1 wk after application. Importantly, yield loss does not correlate with visual symptomology but rather with boll load as noted by plant mapping. Drift or contamination occurring between 9-lf and FB+2 wk has the greatest likelihood to reduce the number of fruit per plant which correlates with greatest potential for yield loss. Injury from 2,4-D nearly always influences plant maturity but a delay in maturity does not always result in yield loss but rather the environment, cultivar, and harvest procedures late in the season can often overcome this effect. The results from this study do contrast some of the previous literature. For example, yield loss was reported to be most severe when drift injury occurred during early growth stages, particularly those prior to bloom (Marple et al., 2008; Everitt and Keeling, 2009; Egan et al., 2014). In a meta-analysis incorporating 20 studies on the yield effect from 2,4-D drift in cotton, Egan et al. (2014) reported that yield losses are most severe during the vegetative and squaring stages, while the detrimental effect on yield lessens when cotton is exposed during the later flowering and boll stages. It is unclear why conflicting differences exist in regards to stage of growth sensitivity when comparing results from this research to the literature. However, it is apparent that more work needs to be done in developing a method for evaluating 2,4-D injury on cotton during later, reproductive growth stages and predicting the corresponding effect on yield.

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Table 2.1. Details for locations of experiments in 2013 and 2014.

Location	Year	Longitude	Latitude	Elevation (m)	Soil Type[a]	Planting Date	Harvest Date
Lewiston, NC	2014	36°07’56.85”N	77°10’14.17”W	21.6	Goldsboro sandy loam	May 14	November 14
Moultrie, GA	2013	31°08’24.19”N	83°43’02.24”W	89.9	Leefield loamy sand	April 26	September 19
New Deal, TX	2013	33°44'13.76"N	101°43'58.04”W	996.1	Pullman clay loam	May 15	October 31
New Deal, TX	2014	33°44'13.76"N	101°43'58.04”W	996.1	Pullman clay loam	June 5	November 20
Portageville, MO	2014	36°24’52.86”N	89°42’04.86”W	87.8	Tiptonville silt loam	May 8	October 22
Quincy, FL	2014	30°32’42.61”N	84°35’38.84”W	75.5	Dothan-Fuquay loamy fine sand	June 5	November 3
Snook, TX	2013	30°30'34.27"N	96°25'13.35"W	66.1	Belk clay	May 6	September 12
Snook, TX	2014	30°30'34.27"N	96°25'59.82"W	66.1	Belk clay	April 16	October 17
Starkville, MS	2013	33°28’17.76”N	88°46’58.18”W	89.9	Mantachie loam	May 15	October 18
Starkville, MS	2014	33°28’17.76”N	88°46’58.18”W	89.9	Mantachie loam	May 7	October 23
Tifton, GA	2013	31°26’16.52”N	83°34’86.44”W	98.8	Tifton loamy sand	April 25	October 11
Tifton, GA	2014	31°26’16.52”N	83°34’86.44”W	98.8	Tifton loamy sand	April 28	September 23

[a] Source: USDA NRCS, 2015 (http://websoilsurvey.sc.egov.usda.gov/App/WebSoilSurvey.aspx)

Table 2.2. Cotton injury and yield response to two rates of 2,4-D for location groups I, II, and III.

Growth stage[a]	2,4-D	Maximum injury[b]	Yield
	g ae ha^{-1}	—— % ——	–% of NTC –
Group I[c]			
4-lf	2	61	101
	40	36	52*
9-lf	2	49	101
	40	29	49*
FB	2	12	109
	40	2	72*
FB+2wk	2	7	106
	40	4	84*
FB+ 4wk	2	5	104
	40	3	104
FB+6wk	2	0	107
	40	0	104
PLSD$_{0.05}$		9	21
Group II			
4-lf	2	74	97
	40	48	66*
9-lf	2	63	94
	40	50	54*
FB	2	41	81*
	40	23	19*
FB+2wk	2	24	94
	40	5	34*
FB+4wk	2	18	102
	40	5	81*
FB+6wk	2	8	107
	40	1	91
PLSD$_{0.05}$		7	17
Group III			
4-lf	2	75	80*
	40	45	48*
9-lf	2	77	77*
	40	56	23*
FB	2	45	60*
	40	21	21*
FB+2wk	2	18	76*
	40	5	47*
FB+4wk	2	20	84
	40	4	67*
FB+6wk	2	13	96
	40	2	84
PLSD$_{0.05}$		8	13

[a] Application timings by growth stage include the four leaf (4-lf), nine leaf (9-lf), first bloom (FB), two weeks after first bloom (FB+2wk), four weeks after first bloom (FB+4wk), and six weeks after first bloom (FB+6wk).

[b] Injury scale of 0 (no injury) to 100% (plant death).

[c] Group I includes New Deal, TX 2013, Snook, TX 2013, Starkville, MS 2014 (yield only), and New Deal, TX 2014 locations; Group II includes Tifton, GA 2013, Tifton, GA 2014, Quincy, FL 2014 (yield only), and Snook, TX 2014 locations; Group III includes Moultrie, GA 2013, Starkville, MS 2013 (yield only), Portageville, MO 2014 (yield only), and Lewiston, NC 2014 locations.

*Denotes significant difference within group at α 0.05. Yield percentage compared to non-treated yield (100%).

Table 2.3. Plant mapping results of total bolls and percent of open bolls at all three location groups.

Growth Stage[a]	2,4-D	Group I[b]		Group II		Group III	
	g ae ha^{-1}	—Total Boll no.—	—% Open—	—Total Boll no.—	—% Open—	—Total Boll no.—	—% Open—
NTC		6.8	94	13.0	90	12.2	75
4-Lf	2	6.7	80	12.3	76	12.9	73
	40	5.0	61	11.0	48	10.3	49
9-Lf	2	6.7	91	13.1	77	11.2	48
	40	4.6	65	9.7	46	7.3	32
FB	2	6.9	87	11.9	75	8.0	80
	40	5.0	81	6.7	62	4.8	80
FB+2wk	2	6.0	94	11.9	85	10.1	81
	40	5.0	92	7.6	87	8.3	79
FB+4wk	2	6.7	92	11.0	89	9.8	79
	40	7.0	91	11.0	91	10.4	80
FB+6wk	2	6.7	92	12.6	84	12.3	77
	40	6.8	91	12.3	84	12.6	77
PLSD$_{0.05}$		1.8	13	3.2	15	3.3	20

[a] Application timings by growth stage include the four leaf (4-lf), nine leaf (9-lf), first bloom (FB), two weeks after first bloom (FB+2wk), four weeks after first bloom (FB+4wk), and six weeks after first bloom (FB+6wk).

[b] Group I includes New Deal, TX 2013, Snook, TX 2013, Starkville, MS 2014, and New Deal, TX 2014 locations; Group II includes Tifton, GA 2013, Tifton, GA 2014, Quincy, FL 2014, and Snook, TX 2014 locations; Group III includes Moultrie, GA 2013, Portageville, MO 2014, and Lewiston, NC 2014 locations.

Figure 2.1. Cotton fruit injury as a result of 2,4-D application. Fruit injury was accompanied by no other visual symptoms of 2,4-D damage.

CHAPTER 3

CHLOROPHYLL *a* FLUORESCENCE PARAMETERS ARE NOT PREDICTIVE OF SUB-LETHAL 2,4-D INDUCED INJURY OR YIELD LOSS IN COTTON (*GOSSYPIUM HIRSUTUM*)[1]

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[1] S.A. Byrd, J.L. Snider, A.S. Culpepper, J.R. Whitaker, P.M. Roberts, W.M. Porter, and G.D. Collins. To be submitted to *Journal of Agronomy and Crop Science*.

Abstract

The commercialization of 2,4-D resistant crops has heightened attention to the potential impact of 2,4-D on non-target sensitive vegetation, as the acreage on which this herbicide is utilized is likely to rise. Previous studies have shown the utility of fluorescence parameters to detect the physiological impact of a multitude of herbicides on various plants. However, this technique has not been evaluated as a potential tool to help determine the impact of a sub-lethal rate of herbicide to a crop. The objective of this study was to quantify the effect of sub-lethal rates of 2,4-D on susceptible cotton with novel chlorophyll *a* fluorescence parameters. An additional objective was to determine if the use of novel chlorophyll *a* fluorescence parameters could be utilized to predict yield loss of cotton exposed to sub-lethal rates of 2,4-D at various growth stages. Treatments included a factorial of two rates of 2,4-D and six cotton growth stages. A non-treated control was included for comparison. Chlorophyll *a* fluorescence parameters including maximum quantum yield of PS II, quantum yield of electron transport, and photosynthetic performance index were measured weekly on all treatments. While researchers have shown this technique to be a valuable tool in documenting plant injury from other herbicides, especially PS II inhibiting herbicides, it is not a viable method to document effects of sub-lethal rates of 2,4-D on cotton. In the present study, cotton exposed to 2,4-D exhibiting significant visual injury and yield losses often had greater photosynthetic efficiency than the non-treated. Thus, this approach will not effectively predict injury or yield response of non-tolerant cotton exposed to 2,4-D at sub-lethal rates.

Introduction

A popular choice for decades for the control of broadleaf weeds (Peterson, 1967), 2,4-dichlorophenoxyacetic acid (2,4-D) is prone to off-target movement, particularly with certain formulations, resulting in drift damage to non-target crops and other surrounding vegetation (Robinson and Fox, 1978; Bayley et al., 1992). 2,4-D and synthetic auxin herbicides in general, function by mimicking the auxin hormone in the plant cell, resulting in a hormonal imbalance (Grossman, 2000; Hansen and Grossman, 2000; Grossman, 2010; Mithila et al., 2011). In response to this imbalance, hormones and compounds are synthesized and released to regulate growth (Grossman et al., 2001; Grossman, 2010). These hormones and compounds, including ethylene, cyanide, and abscisic acid, accumulate and reach toxic levels, resulting in unregulated plant growth, inhibition of physiological processes evidenced by necrosis and senescence of plant tissues, and ultimately plant death (Grossman, 2000; Hansen and Grossman, 2000; Romero-Puertas et al., 2004; Grossman, 2010; Mithila et al., 2011).

The ester and amine formulations of 2,4-D have been reported to be highly susceptible to drift during application, specifically the more volatile ester formulation (Grover et al., 1972; Farwell et al., 1976; Robison and Fox, 1978). Drift from spray droplets or volatility has been an issue of concern with 2,4-D, which has been observed to travel several kilometers from the site of application (Farwell et al., 1976), including a report of 2,4-D drifting over 32 km (Robinson and Fox, 1978). Cotton is one of the most sensitive crops in terms of damage and yield loss from drift, or sub-lethal rates of 2,4-D (Robinson and Fox, 1978; Bayley et al., 1992; Lyon et al., 1993). Leaf and stem malformation, the abortion of fruiting structures, and yield loss have been observed in cotton exposed to sub-lethal rates of 2,4-D in previous simulated drift studies (Marple et al., 2007; 2008; Everitt and Keeling, 2009; Johnson et al., 2012). Consequently, technologies of 2,4-D resistant cotton (Enlist™, Dow AgroSciences, Indianapolis, IN) are nearing commercial release and will enable post-emergence applications of 2,4-D (Dow AgroSciences, 2012), with a new formulation of 2,4-D engineered to be less prone to drift (Dow

AgroSciences, 2011). However, an interface will exist between the new 2,4-D resistant cultivars and susceptible cultivars, increasing the potential for 2,4-D drift damage on susceptible cotton at a multitude of growth stages. Given the increased usage of 2,4-D, as well as the sensitivity of cotton to the herbicide, the potential exists for significant yield losses to result from drift.

Visual rating of injury symptoms have been the typical method for quantifying damage from cotton exposed to 2,4-D drift (Sciumbato et al., 2004; Marple et al., 2007; 2008; Everitt and Keeling, 2009; Johnson et al., 2012); however, visual ratings of injury are not consistently correlated with yield loss (Johnson et al., 2012). The use of aerial video and digital imaging has also been evaluated as an indicator or predictor of herbicide injury; however, the results suggested that this method was less accurate than visual estimates (Hickman et al., 1991).

Previous studies have utilized parameters of chlorophyll *a* fluorescence to detect herbicidal effects on photosynthetic efficiency. These methods are useful because they are rapid (1 to 2 seconds per sample) and provide information on photosynthetic efficiency of the thylakoid reactions (Maxwell and Johnson, 2000). The maximum quantum yield of photosystem II (F_v/F_m), a measure of the potential efficiency of electron transport through photosystem II (electrons transported per photon of light absorbed), and the actual quantum yield of electron transport through photosystem II (ϕ_{PSII}) are the two most commonly utilized chlorophyll *a* fluorescence parameters in plant physiology (Maxwell and Johnson, 2000). Measurements of F_v/F_m are taken on dark-adapted leaves (leaves that have been in the dark long enough to reach a maximal F_v/F_m), whereas ϕ_{PSII} measurements are taken on leaves exposed to a known light level long enough for ϕ_{PSII} to reach stability. Fluorescence parameters have shown utility in previous studies in detecting the effects of a variety of herbicides, on aquatic grasses and algae (Haynes et al., 2000; Ralph, 2000; Eullaffroy and Vernet, 2003). However, few studies have utilized these parameters for the detection of herbicide impact in agronomic plant species, and in particular the effect of 2,4-D on these parameters in cotton appears to be largely unexplored. Although much of the previous work has utilized chlorophyll *a* parameters to detect the effects of PS I or PS II inhibiting herbicides such as paraquat, atrazine, diuron, metribuzin, and simazine (Haynes et al., 2000; Ralph, 2000; Eullaffroy and Vernet, 2003), these parameters may reflect the decrease in photosynthetic rates, damage to chloroplasts, and reduced stomatal aperature resulting from 2,4-D (Grossman et al., 1996; Romero-Puertas, 2004; Grossman, 2010).

Relatively recent, high-resolution fluorescence methods that provide a kinetic description of the rise in fluorescence intensity from F_o (fluorescence level of dark-adapted leaves prior to exposure to a saturating light intensity) to F_m (maximum fluorescence intensity immediately following exposure to a saturating flash of light) allow researchers to estimate typical fluorescence parameters (i.e. F_v/F_m) in addition to some more novel parameters (Strasser et al., 2000). Specifically, using this new methodology (termed OJIP fluorescence) a single fluorescence pulse is broken down into the following steps: the O step (fluorescence intensity at this step = F_o), the J step (fluorescence intensity at 2 ms of exposure to saturating light), the I step (fluorescence at 30 ms exposure), and the P step (F_m regardless of time required to reach maximal fluorescence). OJIP-based methods have become widely used for quantifying photosynthetic properties, particularly because of the wide array of photosynthetic parameters that can be derived from a single measurement, and the short amount of time needed to measure a large number of samples (Appenroth et al., 2001; Strasser et al., 2004; Strauss et al., 2006; Yusuf et al., 2010; Stirbet and Govindjee, 2011). Previous studies have utilized OJIP analysis to measure structural or functional changes in photosynthetic performance (Stirbet and Govindjee, 2011), or plant vitality (Strasser et al., 2004; Oukarroum et al., 2007) in response to various factors (Srivastava et al., 1997; Srivastava et al., 1998; Srivastava et al., 1999; Strasser et al., 2004; Strauss et al., 2006). For a more detailed description of the OJIP methodology and the various parameters measured refer to Stirbet and Govindjee (2011), Strasser et al. (2004), and Strasser et al. (2000). While both F_v/F_m and ϕ_{EO} (roughly equivalent to Φ_{PSII} but measured in dark-adapted leaves) are included in the OJIP analysis, a third parameter, the photosynthetic performance index (PI_{ABS}) (Strasser et al., 2004; Strauss et al., 2006; Oukarroum et al., 2007; Stirbet and Govindjee, 2011) shows the greatest potential utility for plant stress detection, as it has previously been implemented as a screening tool for chilling, drought, and heat tolerance in multiple plant species (Strauss et al., 2006; Oukarroum et al., 2007, 2009) and has been correlated with biomass production in beech trees (Clark et al., 2000).

Because chlorophyll *a* fluorescence is a rapid method for quantifying photosynthetic efficiency in plants, fluorescence is a prime candidate not only for the determination of 2,4-D injury to cotton, but these measurements could also potentially be correlated with yield loss to provide growth-stage specific fluorescence thresholds at which a given level of yield loss could be expected. The objectives of this study were to quantify the effect of sub-lethal rates of 2,4-D on chlorophyll *a* fluorescence responses (F_v/F_m, ϕ_{EO}, and PI_{ABS}) of cotton at four different growth stages. An additional objective of this study was also to determine if any of these physiological parameters could be correlated to 2,4-D induced yield reductions. We hypothesized that if any of these parameters could be identified as accurate predictors of yield loss, with the same or better accuracy compared to the visual estimates that have been used in the past, then the potential exists for chlorophyll fluorescence measurements to quantify injury and estimate yield loss due to 2,4-D injury in cotton.

Materials and Methods

Field experiments were conducted during 2013 at two experimental locations: University of Georgia Gibbs Farm in Tifton, GA (31.26°N, 83.35°W) and the Sunbelt Agricultural Exposition in Moultrie, GA (31.08°N, 83.43°W). The soil type at the Tifton location was Tifton loamy sand (USDA NRCS 2014a), classified as fine-loamy, kaolinitic, thermic Plinthic Kandiudults and being very deep, well drained, with moderately slow permeability (USDA NRCS 2014b). The soil type at the Moultrie location was a Leefield loamy sand (USDA NRCS 2014a), classified as loamy, siliceous, subactive, thermic Arenic Plinthaquic Paleudults and being very deep, somewhat poorly drained, with moderately slow to slow permeability (USDA NRCS 2014b). The cultivar PhytoGen™ 499 WRF (PHY 499) was planted at a rate of 11.5 seeds m^{-1} in both locations on 25 April in Tifton and 1 May in Moultrie. Plots consisted of four rows 91.5 cm in width and 13.7 m in length. The center two rows served as treatment rows on which 2,4-D was applied and all data was taken, with the outer two rows serving as boarders between plots. All crop management practices followed extension recommendations (Collins et al., 2015).

Two sub-lethal rates of Weedar 64 (Nufarm Americas Inc. Alsip, IL, USA), an amine formulation of 2,4-D, representing fractions of the full rate (0.532 kg ae ha^{-1}) were applied through a CO_2 backpack sprayer. One rate, representing a drift scenario, applied 2 g ae ha^{-1} (drift rate) equivalent to 1/421 of the full rate while the second rate represented a tank contamination scenario and applied 40 g ae ha^{1} (contamination rate) representing 1/21 of the full rate. The CO_2 backpack sprayer was calibrated to deliver 140.3 liters per hectare at a nozzle pressure of 1.83 kg cm^{-2}.

To determine the impact of 2,4-D at different growth stages, a single application of both aforementioned rates were made to cotton at four different growth stages, including the four-leaf (4-lf) stage, nine-leaf (9-lf) stage, first bloom (FB), and two weeks after first bloom (FB+2 wk), and were compared to a non-treated control. A factorial arrangement of treatments was utilized and replicated four times at both locations.

The parameters F_v/F_m, ϕ_{EO}, and PI_{ABS} were derived from fluorescence transients obtained on dark-adapted leaves at the fourth main stem node below the apical meristem using a FluorPen™ FP 100-Max fluorometer (Photon Systems Instruments, Brno, Czech Republic) and the OJIP protocol of this particular instrument. Detailed explanations of the mathematical derivation of each of the aforementioned parameters from the chlorophyll *a* fluorescence transient have been given in Strasser et al. (2000) and Strauss et al. (2006). To ensure leaves were dark-adapted, the week prior to the first application, measurements were taken at five minute intervals, beginning at sundown until the readings were consistent, to determine when F_v/F_m was at a maximum and stable. Based on this preliminary data (data not shown), readings were then taken weekly seven days after the four leaf application, beginning at 22:00 hours, until the end of the bloom period. During each measurement period three readings were collected on three plants in each plot, regardless of 2,4-D application.

Cotton was mechanically harvested using a John Deere 9930 (John Deere, Moline, IL) two-row spindle picker with a bagging attachment so that the entire 13.7 m length of both treatment rows could be picked simultaneously. Seedcotton weights were obtained after harvest using Intercomp CS750 digital scale (Intercomp Company, Medina, MN).

Treatment means for all three parameters measured (random effects) were determined for each rate and growth stage (fixed effects) and compared to the control using analysis of variance, with means separated by least significant difference at $\alpha = 0.05$ using SAS 9.4 software (SAS Institute Inc., Cary, NC). Locations were analyzed separately due to the yield effects from 2,4-D. Within each location, growth stage was also analyzed independently due to differences in yield with the date of measurement separated and compared to the control values measured on the same day.

Regressions of plot means for all three fluorescence-based parameters measured versus yield loss compared to control, were performed utilizing JMP Pro 11 (SAS Institute Inc., Cary, NC), with locations analyzed separately. At each location means from each fluorescence parameter were pooled across simulated 2,4-D rate treatments for each growth stage treatment.

Results and Discussion

Data collection dates relative to treatment applications for each growth stage are presented in Table 3.1. Only measurements taken within 50 days of each 2,4-D application are presented to capture the entirety of symptoms and the initiation of recovery. When data were pooled over a collection period, or during the same days after application (DAA) range, growth stage was significant for all parameters with the exception of PI_{ABS} at two weeks after application in Tifton. Thus, growth stages were analyzed separately. When observing the effects of sub-lethal rates of 2,4-D on OJIP-derived parameters, significant differences were present within each growth stage among the different 2,4-D rates, thus rates were analyzed independently. Analysis of variance results for the effect of 2,4-D rate at both locations for all measurement periods are presented in Table 3.2.

Tifton

The range of quantum yield of PS II (F_v/F_m), quantum yield of electron transport (ϕ_{EO}), and photosynthetic performance index (PI_{ABS}) vales in the control for all measurement dates in Tifton are given in Table 3.3.

Four Leaf Applications. An unadvised application of granular nitrogen fertilizer to wet leaves early in the morning lead to leaf desiccation which is reflected in the 21 DAA measurements in the 4-Lf stage (Table 3). The effects of fertilizer desiccation were more evident in control plants, as leaf malformation due to application of 2,4-D resulted in less leaf area exposed to fertilizer granules. Significant differences were present for F_v/F_m values at the four leaf stage in two out of the five measurement periods (Table 3.2). Compared to the control, F_v/F_m was decreased by 2% resulting from the contamination rate at seven DAA and by 1% in both rates of 2,4-D at 30 DAA (Table 3.4). At 21 DAA the contamination rate resulted in a 37 and 33% increase in ϕ_{EO} over the control and drift rate, respectively. The contamination rate increased PI_{ABS} at 21 DAA by 115% over the control and 94% over the drift rate. When compared to the range of control values, the only instance in which a parameter measured was out of this range was ϕ_{EO} in the contamination rate at 21 DAA, which was slightly higher (0.55) than the maximum control value (0.54) on this date.

Nine Leaf Applications. The effect of fertilizer desiccation is also reflected in the seven DAA measurements for the 9-Lf stage, which was the same date as the 21 DAA in the 4-lf, in Tifton, GA (Table 3.5). At 26 DAA, F_v/F_m in the control was reduced by 1% compared to both 2,4-D rates. The drift rate of 2,4-D resulted in a 32% increase in ϕ_{EO} compared to the control at seven DAA. The drift rate also increased PI_{ABS} at 26 to 70% compared to the control during the first three measurements, while the contamination rate resulted in a 41% increase in PI_{ABS} at 26 DAA. In two instances of a significant effect on PI_{ABS} due to 2,4-D, the treatment means (6.15

in the drift rate at 16 DAA; 5.02 in the contamination rate at 26 DAA) exceeded the maximum value observed in the range of control values (6.03 and 4.84 at 16 and 26 DAA, respectively).

First Bloom Applications. Applications made at the FB stage resulted in no instances of significant differences among the 2,4-D rates and control for any of the parameters (Table 3.6).

Two Weeks After First Bloom Application. Applications made at the FB+2 wk stage also resulted in no instances of significant differences among the 2,4-D rates and control for any of the parameters (Table 3.7).

Seasonal patterns of F_v/F_m for each growth stage and 2,4-D rate treatment, including all collection dates, are presented in Figure 3.1A for the Tifton location. The effect of the fertilizer desiccation is evident at the measurements occurring on 19 June. Seasonal patterns of ϕ_{EO} and PI_{ABS} at Tifton are also presented in Figure 3.1B and Figure 3.1C, respectively.

Moultrie

The range of quantum yield of PS II (F_v/F_m), quantum yield of electron transport (ϕ_{EO}), and photosynthetic performance index (PI_{ABS}) in the control for all measurement dates in Moultrie are presented in Table 3.8.

Four Leaf Applications. A difference in F_v/F_m was present between the two 2,4-D rates at 30 DAA, as the contamination rate resulted in a 2% decrease in F_v/F_m compared to the drift rate (Table 3.9). At 49 DAA the contamination rate resulted in a 6% decrease in F_v/F_m compared to both the control and drift rate. Values of ϕ_{EO} at 21 DAA were higher in both the sub-lethal 2,4-D rates than the control, with 11 and 8% increases resulting from the drift and contamination rates, respectively. PI_{ABS} values were 37% higher in the contamination rate compared to the drift rate at 7 DAA, while the drift rate resulted in a 67% increase in PI_{ABS} over the control at 21 DAA. The F_v/F_m value measured at 49 DAA in the contamination rate (0.77) was lower than the minimum control value measured at this date (0.80). The effect of both rates of 2,4-D on PI_{ABS} at 21 DAA (8.0 in the drift rate and 6.4 in the contamination rate) exceeded the maximum range of control values observed on that date (5.99).

Nine Leaf Applications. There was no effect of 2,4-D on F_v/F_m at any of the measurement dates (Table 3.10). The contamination rate resulted in a 6% increase in ϕ_{EO} compared to the control, and a 24% increase in PI_{ABS} over the drift rate at six DAA, though these values did not exceed the range of control values observed.

First Bloom Applications. 2,4-D had no effect on F_v/F_m at any of the measurement dates for applications made at FB (Table 3.11). At 31 DAA there was a 4 and 6% increase in ϕ_{EO} over the control in the drift and contamination rates, respectively. PI_{ABS} was increased by 30% at 14 DAA and 29% at 31 DAA compared to the control. However, the instances of significant differences in chlorophyll *a* fluorescence parameters did not produce values outside the range of control values measured on the respective dates.

Two Weeks after First Bloom Applications. The only instance of 2,4-D effect resulted in a 1% increase in F_v/F_m in the drift rate compared to F_v/F_m in both the control and contamination rate (Table 3.12). The F_v/F_m values recorded in the drift rate on this date (0.85) were also higher than the maximum range of control values (0.84).

Seasonal patterns for F_v/F_m, ϕ_{EO}, and PI_{ABS} are shown in Figures 3.2A, 3.2B, and 3.2C, respectively, for all measurement dates at the Moultrie location.

Injury Ratings. Visual injury ratings of leaf malformation and stem epinasty, on a scale from 0% (no injury) to 100% (plant death), were taken throughout the season at both Tifton and Moultrie locations. There were seven instances across all growth stages in Tifton at which injury evaluations were performed during the same week as chlorophyll *a* fluorescence measurements were also taken. At five of these dates, there were no significant differences present in F_v/F_m, ϕ_{EO}, or PI_{ABS} due to 2,4-D applications, with visual injury ranging from 2 to 60% in the drift rate treatment, and 17% to 59% in the contamination rate (data not shown). At 26 DAA in the 9-lf treatment, when 30 and 60% injury was observed in the drift and contamination treatments, respectively, F_v/F_m and PI_{ABS} were lower in the control treatment than either treatment receiving a 2,4-D application. At 30 DAA in the 4-lf stage in Tifton, the one instance in which an influence from 2,4-D was observed in both injury ratings and chlorophyll *a* fluorescence, F_v/F_m was reduced in both 2,4-D rates compared to the control. This was followed by injury ratings at 34 DAA of 41 and 71% in the drift and contamination rates, respectively, the highest injury ratings in the 4-lf treatment during the season. However, the decrease in actual difference in F_v/F_m (0.84 in the control, 0.83 in both 2,4-D rates) was relatively small and likely not reflective of a biological effect of 2,4-D on the quantum yield of PS II, or the visual symptoms of injury. Further, 0.83 is considered the maximum F_v/F_m value for most species (Maxwell and Johnson, 2000), while the contamination rate at the 4-lf stage in Tifton resulted in significant yield loss compared to the control (Fig. 3.3A).

There were 11 instances in Moultrie in which injury ratings were taken within the same week as chlorophyll *a* fluorescence measurements. Injury across all growth stages ranged from 1 to 60% in the drift rate and 18 to 75% in the contamination rate. However, over all 11 dates, there was only one instance of a negative impact on F_v/F_m, ϕ_{EO}, or PI_{ABS} observed through 2,4-D applications, with F_v/F_m being lower in the contamination rate compared to the control at 49 DAA in the 4-lf treatment, a treatment which resulted in yield loss compared to the control (Fig. 3.3B) Injury ratings taken in the 4-lf treatment were 8 and 45% at 44 DAA in the drift and contamination rate, respectively. At both locations, results of chlorophyll *a* fluorescence did not mirror the effect of 2,4-D rate or growth stage on visual injury evaluations, as values of these parameters did not reach levels considered to be indicative of inhibited photosynthetic performance

Seedcotton Yield Loss Regressions. To determine the magnitude of yield loss compared to the control on a percentage basis, the yield of each treatment was converted to the percent of control yield produced. The contamination rate of 2,4-D resulted in significant yield loss, compared to the control, at all growth stages at both Tifton (Fig. 3.3A) and Moultrie (Fig. 3.3B). The drift rate resulted in loss when applied at the FB stage in Tifton, and at the 9-lf, FB, and FB+2 wk stages at Moultrie. To determine the relationship between F_v/F_m, ϕ_{EO}, or PI_{ABS} on seedcotton yield, the actual difference in yield, on a kg ha^{-1} basis, was determined for each treatment to determine yield reductions resulting from the drift and contamination rate of 2,4-D at the four growth stages. Yield reductions for each 2,4-D rate and growth stage application were determined by comparing the yield of each growth stage treatment (with the yield pooled across 2,4-D rates at each growth stage) to the yield of the control. Yield loss, compared to the control at the Tifton location ranged from 1790.2 kg/ha to 616 kg ha^{-1} of seedcotton across all growth stages and 2,4-D rates. In Moultrie, yield loss compared to the control ranged from 2370 kg/ha to 935.8 kg/ha across all growth stages and 2,4-D rates. The results of the regression analysis of parameter means by yield loss for each treatment at the Tifton location are presented in Table 3.13. Over the course of the season and all data collection periods, only two instances of significance at this location occurred. A significant negative relationship between yield loss and F_v/F_m occurred at 14 DAA in the 4-Lf growth stage (Fig. 3.4A), suggesting that greater yield loss, compared to the control, occurred from the applications of 2,4-D rates at this growth stage. In contrast, a significant positive relationship between these two parameters was observed when measurements were taken at six DAA for the FB application (Fig. 4B). There was only one instance of significance in the yield loss regressions performed on fluorescence-derived data collected at the Moultrie location (only for F_v/F_m) (Table 3.14). In this case, it occurred in the FB+2wk growth stage at 22 DAA, with the direction of the effect being positive (Figure 4C).

Previous studies on the effects of herbicides on photosynthetic or fluorescence parameters have seen a significant result on species of algae and aquatic grasses; however, it is critical to note that the majority of these studies included herbicides that directly inhibit photosystem II (Haynes et al., 2000; Ralph, 2000; Eullaffroy and Vernet, 2003). In each of these studies, photosystem II inhibiting herbicides negatively impacted F_v/F_m while herbicides with other modes of action, such as glyphosate, were determined to have no effect, even at rates 100-times greater than the photosystem II inhibiting herbicides (Ralph, 2000). In instances when 2,4-D was included, Wong (2000) reported that the herbicide had an effect on photosynthetic rates of algae; however, it appears that the effect of 2,4-D on fluorescence responses in higher plants is largely unexplored, especially regarding the impact of this herbicide on fluorescence transients of cotton.

Across the four growth stages in this study, there were 18 total collection dates of each parameter at Tifton and 16 at Moultrie. Across both locations and growth stages, F_v/F_m was significantly different from the control in 6 instances out of a possible 34. In many cases, while the differences were statistically significant, it is doubtful that the 2,4-D resulted in any negative impacts of biological relevance on F_v/F_m. For this technique to be useful for determining 2,4-D injury leading to yield loss, lower F_v/F_m values would need to be consistently observed in 2,4-D treated plants relative to control plants. In contrast, at 30 DAA in the 4-lf treatment in Tifton (Table 3.3), a statistically higher F_v/F_m value was observed in the control (0.839) compared to both the drift and contamination rates of 2,4-D (0.827 and 0.826 respectively). This provides evidence that no predictable, deleterious effect of 2,4-D on F_v/F_m levels was observed at the sub-lethal rates used in the current study, despite the visual observations of harmful effects on plant growth and the resulting yield loss. Furthermore, in the current study, even when statistical differences between simulated 2,4-D rates and the control were observed, F_v/F_m values were still indicative of healthy plants. For example, in cases of statistical significance across all growth stage treatments, the percent difference in F_v/F_m between the treatments ranged from 1.5 to 2.4% in Tifton (with the exclusion of the two dates impacted by fertilizer desiccation) and from 0.8 to 6.3% in Moultrie. While little research has been conducted in cotton on the effect of 2,4-D or other synthetic auxin herbicides on F_v/F_m, previous studies have shown that F_v/F_m values in non-stressed cotton are ≥ 0.8 (Pettigrew, 2004; Burke, 2007; Massacci et al., 2008; Snider et al., 2013). Levels of F_v/F_m in the current study never decreased to levels that would provide evidence that F_v/F_m was inhibited as a result of 2,4-D exposure (mean values for all treatments being higher than 0.8) as would be required for injury and/or yield loss prediction. Although there are instances of significant treatment effects, this may be due to in field variability or some other factor, as there is no evidence that the applications of 2,4-D at sub-lethal rates had a detrimental effect on F_v/F_m.

Significant differences between the ϕ_{EO} values of the 2,4-D rates and the control across both locations and all growth stages were found in only 5 of the possible 34 instances. However, in all five cases of significant difference among treatments, the control treatment had significantly lower ϕ_{EO} values than either one or both of the sub-lethal rates of 2,4-D. This was the case in three out of five instances in Moultrie. In Tifton, the readings that resulted in significant differences (21 DAA in the 4-lf treatment and seven DAA in the 9-lf treatment) were both impacted by fertilizer desiccation (Table 3.3 and 3.4, respectively). In Moultrie, the effect resulted in an 11.1% difference in the 4-lf treatment, a 6.5% difference in the 9-lf treatment, and a 5.7% difference in the FB treatment. Due to the relatively small percent difference between treatments, and the fact that higher values were observed in treatments receiving applications of 2,4-D, it appears that this parameter did not provide evidence of decreased plant function, and would not be a good indicator of a biologically significant impact from 2,4-D of plant injury or yield loss.

Significant differences in the values of PI_{ABS} among 2,4-D treatments across all growth stages and both locations occurred in 9 out of the possible 34 instances. Excluding the two instances impacted by fertilizer desiccation, (in which the parameter was significantly lower in the control than at least one of the 2,4-D treatments) in five of the seven remaining cases of significance, the control presented a significantly lower PI_{ABS} value than either one or both of the 2,4-D rates. One instance of this is illustrated in the 9-lf growth stage in Tifton, where the control PI_{ABS} value was significantly lower than both the drift and contamination rates of 2,4-D (Table 3.4). In terms of the percent difference among treatments where significance was observed (excluding the two measurement dates impacted by the fertilizer desiccation), this ranged from 32.4 to 41.2% in the 9-lf growth stage in Tifton. In Moultrie, the percent difference ranged from 36.7 to 67.4% in the 4-lf growth stage, a 24% difference in the one instance in the 9-lf growth stage, and 29.4 to 30.1% in the FB growth stage. The difference between treatments was greater when the PI_{ABS} parameter was utilized when in comparison to the aforementioned, fluorescence-derived parameters used in this study. However, the fact that the control treatment often had significantly lower PI_{ABS} values compared to cotton exposed to sub-lethal rates of 2,4-D, when yield loss occurred as the result of 2,4-D application, supplies little evidence that this could be a reliable parameter to predict herbicide injury when sub-lethal rates of 2,4-D have been applied.

The incidence of injury to cotton from sub-lethal rates of 2,4-D will inevitably increase with the release of 2,4-D tolerant cotton cultivars, thereby increasing in the interface between tolerant and non-tolerant cultivars. If a method could be utilized to detect 2,4-D injury and give an indicator of expected yield loss, or be correlated to visual injury symptoms, it could become a valuable, objective tool for determining injury severity in terms of expected yield loss and could be used as a decision tool for crop management. Although OJIP analysis of the chlorophyll fluorescence trace is successfully used to detect a number of different stresses in multiple plant species, it appears that this is the first instance in which OJIP analysis has been utilized to detect injury on cotton from exposure to 2,4-D. The results of the current study indicate that OJIP analysis would not be an accurate predictor of injury or corresponding yield loss due to sub-lethal rates of 2,4-D. The lack of consistent patterns in, and overall instances of significant declines in photochemical parameters among the growth stages and 2,4-D treatments would seem to eliminate this method of plant based measurement from being used as an indicator of injury or predictor of yield loss stemming from exposure to sub-lethal rates of 2,4-D. Furthermore, the fact that in the majority of cases the control produced the lowest values across all three fluorescence-based parameters provides evidence that measurements of these processes may not detect 2,4-D injury at the rates used in the current study, even when significant yield loss resulted from 2,4-D, necessitating the exploration of other avenues to detect yield-limiting 2,4-D injury.

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Table 3.1. Measurements dates with days after application (DAA) of 2,4-D for the four growth stage treatments in Tifton and Moultrie.

Measurement No.	Growth Stage Treatment			
	4-lf[\Z]	9-lf	FB	FB+2 wk
Tifton				
1	5 June (7 DAA)	19 June (7 DAA)	8 July (6 DAA)	23 July (6 DAA)
2	12 June (14 DAA)	28 June (16 DAA)	19 July (17 DAA)	31 July (14 DAA)
3	19 June (21 DAA)	8 July (26 DAA)	23 July (21 DAA)	5 August (19 DAA)
4	28 June (30 DAA)	19 July (37 DAA)	31 July (29 DAA)	
5	8 July (40 DAA)	23 July (41 DAA)	5 August (34 DAA)	
Moultrie				
1	4 June (7 DAA)	18 June (6 DAA)	16 July (14 DAA)	25 July (8 DAA)
2	11 June (14 DAA)	27 June (15 DAA)	25 July (23 DAA)	2 August (16 DAA)
3	18 June (21 DAA)	16 July (34 DAA)	2 August (31 DAA)	8 August (22 DAA)
4	27 June (30 DAA)	25 July (43 DAA)	8 August (37 DAA)	
5	16 July (49 DAA)			

[Z] Growth stages at time of application included four leaf (4-lf), nine leaf (9-lf), first bloom (FB), and two weeks after first bloom (FB+2 wk).

Table 3.2. Anova results (F values) for the main effect of 2,4-D rate on fluorescence measurements taken in Tifton and Moultrie, GA. Measurements include the week after 2,4-D was applied.

Tifton, GA	df	Measurement Number				
4-Lf[Z]		1	2	3	4	5
F_v/F_m[Y]	2	6.14*	0.03	3.12	6.02*	1.09
ϕ_{EO}[X]	2	0.56	3.23	5.42*	0.69	2.43
PI_{ABS}[W]	2	0.42	1.53	8.91**	0.92	4.25
9-Lf						
F_v/F_m	2	3.45	4.14	12.02**	1.53	0.98
ϕ_{EO}	2	4.56*	2.45	2.76	0.15	1.34
PI_{ABS}	2	5.32*	6.18*	8.06**	0.02	0.29
FB						
F_v/F_m	2	3.97	0.60	0.90	1.10	0.44
ϕ_{EO}	2	0.92	0.67	3.11	4.07	0.74
PI_{ABS}	2	0.22	0.63	2.87	4.18	0.45
FB+2 wk						
F_v/F_m	2	2.50	0.04	0.54	N/A	N/A
ϕ_{EO}	2	3.52	1.14	0.73	N/A	N/A
PI_{ABS}	2	2.62	0.40	1.01	N/A	N/A
Moultrie, GA						
4-Lf						
F_v/F_m	2	3.48	0.28	2.93	5.77*	9.81**
ϕ_{EO}	2	3.70	0.08	13.59**	4.24	0.78
PI_{ABS}	2	4.90*	0.40	9.80**	0.87	1.51
9-Lf						
F_v/F_m	2	1.34	3.81	1.79	2.53	N/A
ϕ_{EO}	2	7.42*	2.20	0.96	0.93	N/A
PI_{ABS}	2	4.61*	4.84	0.09	0.16	N/A
FB						
F_v/F_m	2	N/A	3.72	0.68	1.84	2.96
ϕ_{EO}	2	N/A	2.07	0.01	13.92**	0.97
PI_{ABS}	2	N/A	6.57*	1.40	6.37*	2.20
FB+2 wk						
F_v/F_m	2	0.97	0.08	7.13*	N/A	N/A
ϕ_{EO}	2	1.11	2.51	0.27	N/A	N/A
PI_{ABS}	2	0.68	0.77	1.45	N/A	N/A

[Z] Applications of sub-lethal 2,4-D rates were made at the four leaf (4-Lf), nine leaf (9-Lf), first bloom (FB) and two weeks after first bloom (FB+2 wk) growth stages.

[Y] F_v/F_m denotes the maximum quantum yield of photosystem II.

[X] ϕ_{EO} denotes quantum yield of electron transport.

[W] PI_{ABS} denotes photosynthetic performance index.

*, ** significant at $\alpha = 0.05$ and 0.01, respectively.

Table 3.3. Range of F_v/F_m, ϕ_{EO}, and PI_{ABS} values in the non-treated control from all measurement dates in Tifton, GA.

Parameter	Growth Stage (DAA)†								
	4-lf (7)	4-lf (14)	4-lf (21) 9-lf (7)	4-lf (30) 9-lf (16)	4-lf (40) 9-lf (26) FB (6)	9-lf (37) FB (17) FB+2 wk (6)	9-lf (41) FB (21) FB+2 wk (14)	FB (29) FB+2 wk (19)	FB (34)
F_v/F_m	0.80 - 0.83	0.81 - 0.84	0.24 - 0.82	0.82 - 0.85	0.80 - 0.83	0.82 - 0.84	0.82 - 0.84	0.83 - 0.85	0.82 - 0.84
ϕ_{EO}	0.53 - 0.59	0.50 - 0.56	0.10 - 0.54	0.49 - 0.56	0.44 - 0.53	0.48 - 0.54	0.50 - 0.54	0.48 - 0.55	0.48 - 0.53
PI_{ABS}	3.81 - 6.92	3.32 - 5.66	0.24 - 4.77	3.14 - 6.03	2.77 - 4.84	3.90 - 5.65	3.79 - 5.35	3.70 - 6.57	3.39 - 5.26

†Growth stages with days after application (DAA) included the four leaf (4-lf), nine leaf (9-lf), first bloom (FB) and two weeks after first bloom (FB+2 wk).

Table 3.4. Results of OJIP fluorescence analysis performed on cotton at the four leaf growth stage at Tifton, GA.

Parameter	2,4-D Rate (g ae ha^{-1})	7 DAA^{ZY}	14 DAA	21 DAA	30 DAA	40 DAA
	0	0.82 a	0.82	0.68	0.84 a	0.81
F_v/F_m^{X}	2	0.82 a	0.82	0.76	0.83 b	0.81
	40	0.80 b	0.82	0.81	0.83 b	0.81
LSD		0.01	NS	NS	0.01	NS
	0	0.56	0.53	0.41 b	0.54	0.51
ϕ_{EO}^{W}	2	0.56	0.53	0.42 b	0.52	0.50
	40	0.55	0.55	0.56 a	0.53	0.53
LSD		NS	NS	0.114	NS	NS
	0	5.28	4.43	2.72 b	4.90	3.55
PI_{ABS}^{V}	2	5.12	4.54	3.02 b	4.24	3.63
	40	4.97	5.14	5.85 a	4.76	4.27
LSD		NS	NS	1.86	NS	NS

^{Z}DAA denotes days after application.
^{Y}Means within a column that share a common letter are not significantly different at $p \leq 0.05$, for each individual parameter.
$^X F_v/F_m$ denotes the maximum quantum yield of photosystem II.
$^W \phi_{EO}$ denotes quantum yield of electron transport.
$^V PI_{ABS}$ denotes photosynthetic performance index.
NS No significant difference.

Table 3.5. Results of OJIP fluorescence analysis performed on cotton at the nine-leaf growth stage at Tifton, GA.

Parameter	2,4-D Rate (g ae ha^{-1})	7 DAA[Z][Y]	16 DAA	26 DAA	37 DAA	41 DAA
	0	0.68	0.84	0.81 b	0.83	0.84
F_v/F_m[X]	2	0.81	0.85	0.82 a	0.83	0.83
	40	0.79	0.84	0.82 a	0.82	0.83
LSD		NS	NS	0.01	NS	NS
	0	0.41 b	0.54	0.51	0.51	0.53
ϕ_{EO}[W]	2	0.54 a	0.55	0.53	0.52	0.54
	40	0.49 ab	0.53	0.53	0.51	0.54
LSD		0.10	NS	NS	NS	NS
	0	2.72 b	4.90 b	3.55 b	4.82	4.83
PI_{ABS}[V]	2	4.61 a	6.15 a	4.70 a	4.82	5.04
	40	3.48 ab	4.65 b	5.02 a	4.72	5.20
LSD		1.32	1.04	0.87	NS	NS

[Z] DAA denotes days after application.

[Y] Means within a column that share a common letter are not significantly different at $p \leq 0.05$, for each individual parameter.

[X] F_v/F_m denotes the maximum quantum yield of photosystem II.

[W] ϕ_{EO} denotes quantum yield of electron transport.

[V] PI_{ABS} denotes photosynthetic performance index.

NS No significant difference.

Table 3.6. Results of OJIP fluorescence analysis performed on cotton at the first bloom growth stage at Tifton, GA.

Parameter	2,4-D Rate (g ae ha^{-1})	6 DAA[Z][Y]	17 DAA	21 DAA	29 DAA	34 DAA
	0	0.81	0.83	0.84	0.84	0.84
F_v/F_m[X]	2	0.82	0.84	0.84	0.84	0.83
	40	0.82	0.83	0.84	0.85	0.83
LSD		NS	NS	NS	NS	NS
	0	0.51	0.51	0.53	0.51	0.53
ϕ_{EO}[W]	2	0.49	0.51	0.53	0.53	0.52
	40	0.50	0.52	0.55	0.55	0.53
LSD		NS	NS	NS	NS	NS
	0	3.55	4.82	4.83	4.88	5.22
PI_{ABS}[V]	2	3.60	5.15	5.06	5.68	4.78
	40	3.50	5.30	5.81	6.57	5.25
LSD		NS	NS	NS	NS	NS

[Z] DAA denotes days after application.

[Y] Means within a column that share a common letter are not significantly different at $p \leq 0.05$, for each individual parameter.

[X] F_v/F_m denotes the maximum quantum yield of photosystem II.

[W] ϕ_{EO} denotes quantum yield of electron transport.

[V] PI_{ABS} denotes photosynthetic performance index.

NS No significant difference.

Table 3.7. Results of OJIP fluorescence analysis performed on cotton at two weeks after first bloom growth stage at Tifton, GA.

Parameter	2,4-D Rate (g ae ha^{-1})	6 DAA^{ZY}	14 DAA	19 DAA
	0	0.84	0.84	0.83
F_v/F_m^{X}	2	0.83	0.84	0.84
	40	0.83	0.84	0.83
LSD		NS	NS	NS
	0	0.53	0.51	0.51
ϕ_{EO}^{W}	2	0.51	0.53	0.52
	40	0.50	0.53	0.50
LSD		NS	NS	NS
	0	4.83	4.88	4.46
PI_{ABS}^{V}	2	4.34	5.34	4.84
	40	3.80	5.28	4.22
LSD		NS	NS	NS

^{Z}DAA denotes days after application.
^{Y}Means within a column that share a common letter are not significantly different at $p \leq 0.05$, for each individual parameter.
^{X}F_v/F_m denotes the maximum quantum yield of photosystem II.
^{W}ϕ_{EO} denotes quantum yield of electron transport.
^{V}PI_{ABS} denotes photosynthetic performance index.
NS No significant difference.

Table 3.8. Range of F_v/F_m, ϕ_{EO}, and PI_{ABS} values in the non-treated control from all measurement dates in Moultrie, GA.

Parameter	Growth Stage (DAA)†							
	4-lf (7)	4-lf (14)	4-lf (21) 9-lf (7)	4-lf (30) 9-lf (15)	4-lf (49) 9-lf (34) FB (14)	9-lf (43) FB (23) FB+2wk (8)	FB (31) FB+2wk (16)	FB (37) FB+2wk (22)
F_v/F_m	0.77 - 0.82	0.71 - 0.83	0.81 - 0.83	0.81 - 0.84	0.80 - 0.84	0.82 - 0.85	0.83 - 0.86	0.83 - 0.84
ϕ_{EO}	0.53 - 0.63	0.37 - 0.59	0.51 - 0.57	0.52 - 0.61	0.49 - 0.59	0.47 - 0.56	0.53 - 0.58	0.52 - 0.56
PI_{ABS}	3.47 - 7.86	2.03 - 6.44	3.65 - 5.99	3.80 - 7.41	3.08 - 6.57	3.34 - 6.07	4.45 - 7.84	4.77 - 6.60

†Growth stages with days after application (DAA) included the four leaf (4-lf), nine leaf (9-lf), first bloom (FB) and two weeks after first bloom (FB+2 wk).

Table 3.9. Results of OJIP fluorescence analysis performed on cotton at the four-leaf growth stage at Moultrie, GA.

Parameter	2,4-D Rate (g ae ha^{-1})	7 DAA^{ZY}	14 DAA	21 DAA	30 DAA	49 DAA
	0	0.81	0.78	0.82	0.82 ab	0.82 a
F_v/F_m^{X}	2	0.79	0.79	0.83	0.82 a	0.82 a
	40	0.81	0.79	0.81	0.80 b	0.77 b
LSD		NS	NS	NS	0.02	0.03
	0	0.58	0.53	0.53 b	0.57	0.54
ϕ_{EO}^{W}	2	0.56	0.53	0.59 a	0.58	0.52
	40	0.61	0.54	0.57 a	0.52	0.52
LSD		NS	NS	0.03	NS	NS
	0	5.97 ab	4.12	4.75 b	5.35	4.84
PI_{ABS}^{V}	2	5.20 b	4.92	7.95 a	6.36	4.50
	40	7.11 a	4.87	6.39 ab	5.03	3.70
LSD		1.39	NS	1.64	NS	NS

^{Z}DAA denotes days after application.

^{Y}Means within a column that share a common letter are not significantly different at $p \leq 0.05$, for each individual parameter.

$^X F_v/F_m$ denotes the maximum quantum yield of photosystem II.

$^W \phi_{EO}$ denotes quantum yield of electron transport.

$^V PI_{ABS}$ denotes photosynthetic performance index.

NS No significant difference.

Table 3.10. Results of OJIP fluorescence analysis performed on cotton at the nine-leaf growth stage at Moultrie, GA.

Parameter	2,4-D Rate (g ae ha^{-1})	6 DAA^{ZY}	15 DAA	34 DAA	43 DAA
	0	0.82	0.82	0.82	0.84
F_v/F_m^{X}	2	0.82	0.83	0.80	0.82
	40	0.83	0.83	0.81	0.82
LSD		NS	NS	NS	NS
	0	0.53 b	0.57	0.54	0.51
ϕ_{EO}^{W}	2	0.52 b	0.60	0.51	0.51
	40	0.56 a	0.59	0.55	0.53
LSD		0.02	NS	NS	NS
	0	4.75 ab	5.35	4.84	4.76
PI_{ABS}^{V}	2	4.39 b	7.66	4.71	4.81
	40	5.44 a	6.27	5.36	5.25
LSD		0.80	NS	NS	NS

^{Z}DAA denotes days after application.

^{Y}Means within a column that share a common letter are not significantly different at $p \leq 0.05$, for each individual parameter.

$^X F_v/F_m$ denotes the maximum quantum yield of photosystem II.

$^W \phi_{EO}$ denotes quantum yield of electron transport.

$^V PI_{ABS}$ denotes photosynthetic performance index.

NS No significant difference.

Table 3.11. Results of OJIP fluorescence analysis performed on cotton at the first bloom growth stage at Moultrie, GA.

Parameter	2,4-D Rate (g ae ha^{-1})	14 DAA^{ZY}	23 DAA	31 DAA	37 DAA
F_v/F_m^{X}	0	0.82	0.84	0.85	0.84
	2	0.83	0.83	0.85	0.84
	40	0.83	0.84	0.85	0.85
LSD		NS	NS	NS	NS
ϕ_{EO}^{W}	0	0.54	0.51	0.55 b	0.54
	2	0.55	0.51	0.57 a	0.55
	40	0.56	0.51	0.58 a	0.57
LSD		NS	NS	0.01	NS
PI_{ABS}^{V}	0	4.89 b	4.76	6.03 b	5.86
	2	5.35 b	4.88	6.83 ab	6.64
	40	6.29 a	5.83	7.80 a	7.74
LSD		0.92	NS	1.12	NS

^{Z}DAA denotes days after application.

^{Y}Means within a column that share a common letter are not significantly different at $p \leq 0.05$, for each individual parameter.

$^X F_v/F_m$ denotes the maximum quantum yield of photosystem II.

$^W \phi_{EO}$ denotes quantum yield of electron transport.

$^V PI_{ABS}$ denotes photosynthetic performance index.

NS No significant difference.

Table 3.12. Results of OJIP fluorescence analysis performed on cotton at two weeks after first bloom growth stage at Moultrie, GA.

Parameter	2,4-D Rate (g ae ha^{-1})	8 DAA^{ZY}	16 DAA	22 DAA
	0	0.84	0.85	0.84 b
F_v/F_m^{X}	2	0.83	0.85	0.85 a
	40	0.83	0.85	0.84 b
LSD		NS	NS	0.01
	0	0.51	0.55	0.54
ϕ_{EO}^{W}	2	0.50	0.54	0.55
	40	0.52	0.56	0.55
LSD		NS	NS	NS
	0	4.76	6.03	5.86
PI_{ABS}^{V}	2	4.22	5.67	6.51
	40	4.37	6.37	5.83
LSD		NS	NS	NS

[Z] DAA denotes days after application.

[Y] Means within a column that share a common letter are not significantly different at $p \leq 0.05$, for each individual parameter.

[X] F_v/F_m denotes the maximum quantum yield of photosystem II.

[W] ϕ_{EO} denotes quantum yield of electron transport.

[V] PI_{ABS} denotes photosynthetic performance index.

NS No significant difference.

Table 3.13. Results of regression analysis (R^2 values) from Tifton, GA.

Parameter	Days After 2,4-D Application				
4-Lf	**7 DAA**^{Z}	**14 DAA**	**21 DAA**	**30 DAA**	**40 DAA**
$F_v/F_m{}^Y$	0.252	0.898*	0.239	0.111	0.210
$\phi_{EO}{}^X$	0.493	0.016	0.174	0.137	0.637
$PI_{ABS}{}^W$	0.564	0.014	0.135	0.376	0.395
9-Lf	**7 DAA**	**16 DAA**	**26 DAA**	**37 DAA**	**41 DAA**
F_v/F_m	0.413	0.632	0.572	0.011	0.173
ϕ_{EO}	0.353	0.471	0.030	0.024	0.120
PI_{ABS}	0.562	0.652	0.029	0.049	0.006
FB	**6 DAA**	**17 DAA**	**21 DAA**	**29 DAA**	**34 DAA**
F_v/F_m	0.693*	0.092	0.203	0.193	0.021
ϕ_{EO}	0.150	0.410	0.128	0.234	0.010
PI_{ABS}	0.127	0.023	0.100	0.168	0.013
FB+2 wk	**6 DAA**	**14 DAA**	**19 DAA**		
F_v/F_m	0.193	0.112	0.249		
ϕ_{EO}	0.013	0.547	0.245		
PI_{ABS}	0.024	0.472	0.447		

^{Z}DAA denotes days after application.

$^Y F_v/F_m$ denotes the maximum quantum yield of photosystem II.

$^X \phi_{EO}$ denotes quantum yield of electron transport.

$^W PI_{ABS}$ denotes photosynthetic performance index.

*R^2 value significant at $p \leq 0.05$.

Table 3.14. Results of regression analysis (R^2 values) from Moultrie, GA.

Parameter	Days After 2,4-D Application				
4 Leaf	**7 DAA^Z**	**14 DAA**	**21 DAA**	**30 DAA**	**49 DAA**
F_v/F_m^{Y}	0.111	0.253	0.44	0.147	0.205
ϕ_{EO}^{X}	0.314	0.071	0.219	0.148	0
PI_{ABS}^{W}	0.232	0.031	0.462	0.015	0.056
9 Leaf	**6 DAA**	**15 DAA**	**34 DAA**	**43 DAA**	
F_v/F_m	0.215	0.359	0.113	0.126	
ϕ_{EO}	0.406	0.321	0.309	0.359	
PI_{ABS}	0.432	0.587	0.08	0.097	
FB	**14 DAA**	**23 DAA**	**31 DAA**	**37 DAA**	
F_v/F_m	0.2	0.175	0.227	0.114	
ϕ_{EO}	0.123	0.025	0.373	0.883	
PI_{ABS}	0.291	0.339	0.286	0.157	
FB+2 wk	**8 DAA**	**16 DAA**	**22 DAA**		
F_v/F_m	0.409	0.088	0.756*		
ϕ_{EO}	0.432	0.127	0.041		
PI_{ABS}	0	0.011	0.455		

[Z] DAA denotes days after application.

[Y] F_v/F_m denotes the maximum quantum yield of photosystem II.

[X] ϕ_{EO} denotes quantum yield of electron transport.

[W] PI_{ABS} denotes photosynthetic performance index.

*R^2 value significant at $p \leq 0.05$.

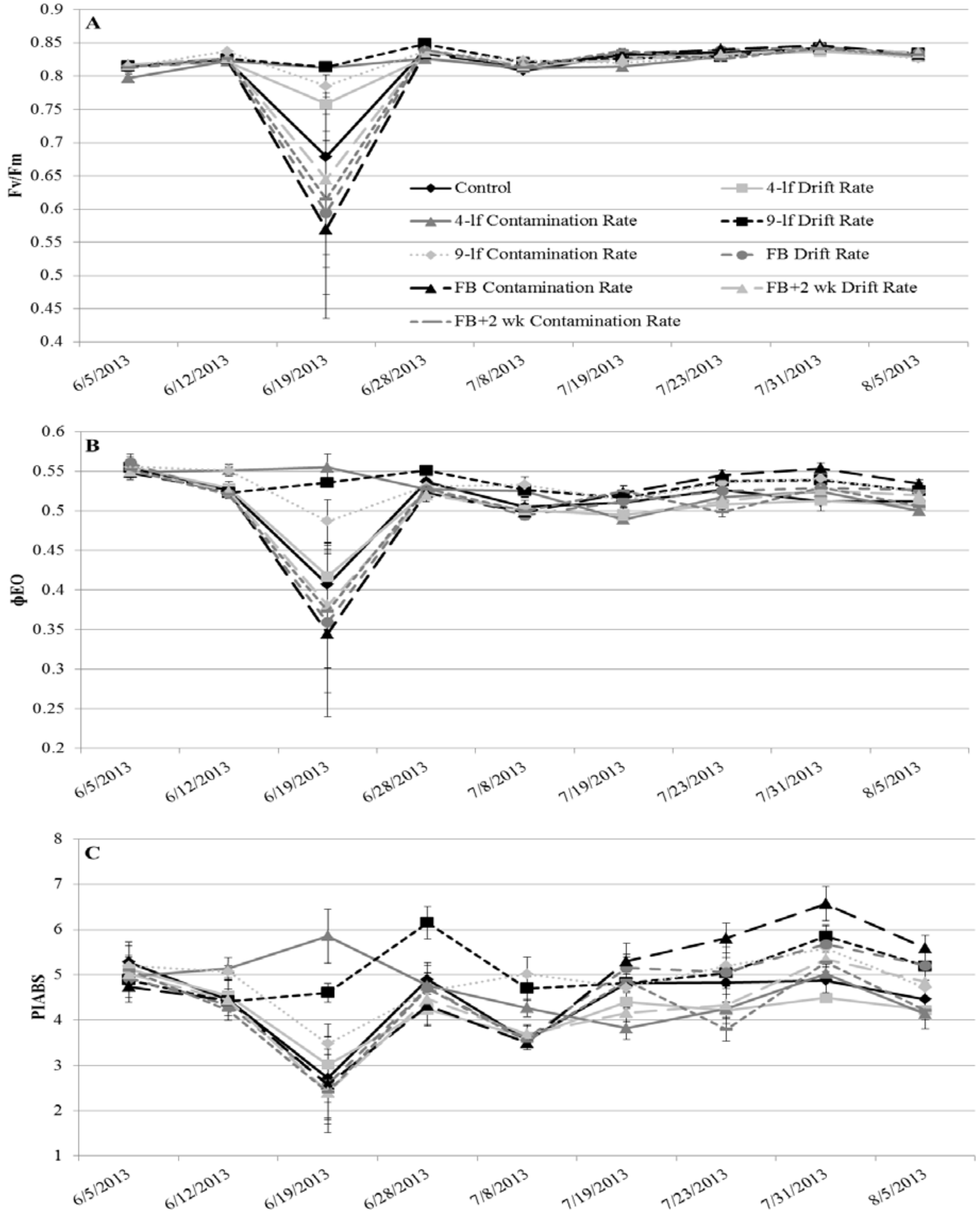

Figure 3.1. Seasonal patterns of F_v/F_m (A) ϕ_{EO} (B), and PI_{ABS} (C) from all treatments and collection dates in Tifton, GA. Growth stage treatments include the four leaf (4-lf), nine leaf (9-lf), first bloom (FB), and two weeks after first bloom (FB+2 wk) stages, with the 2,4-D drift and contamination 2,4-D rates. Applications of 2,4-D were made on the 4-lf treatment on 29 May, on the 9-lf treatment on 12 June, on the FB treatment on 2 July, and on the FB+2 wk treatment on 17 July.

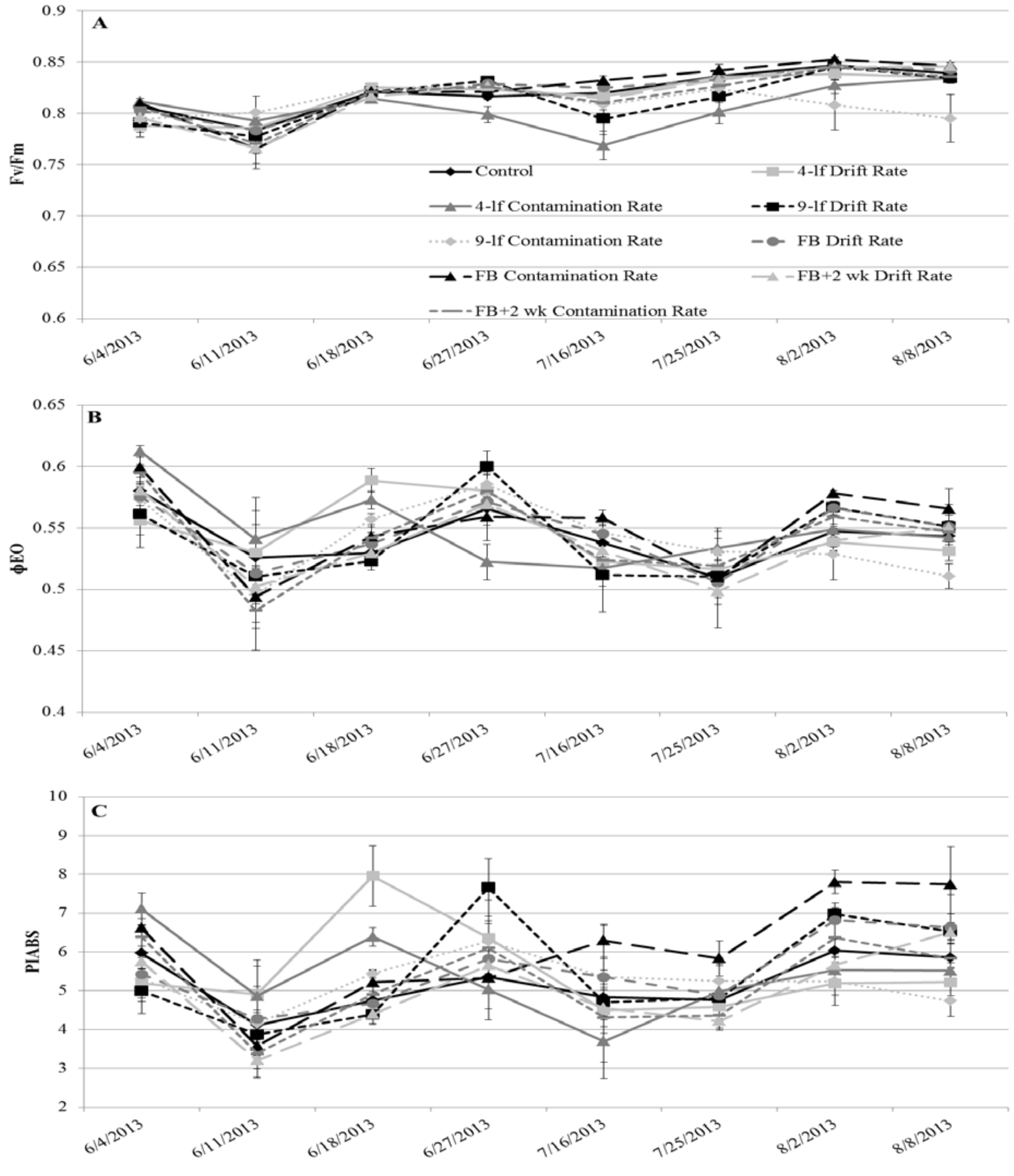

Figure 3.2. Seasonal patterns of F_v/F_m (A) ϕ_{EO} (B), and PI_{ABS} (C) from all treatments and collection dates in Moultrie, GA. Growth stage treatments include the four leaf (4-lf), nine leaf (9-lf), first bloom (FB), and two weeks after first bloom (FB+2 wk) stages, with the 2,4-D drift and contamination rates. Applications of 2,4-D were made on the 4-lf treatment on 28 May, on the 9-lf treatment on 12 June, on the FB treatment on 2 July, and on the FB+2wk on 17 July.

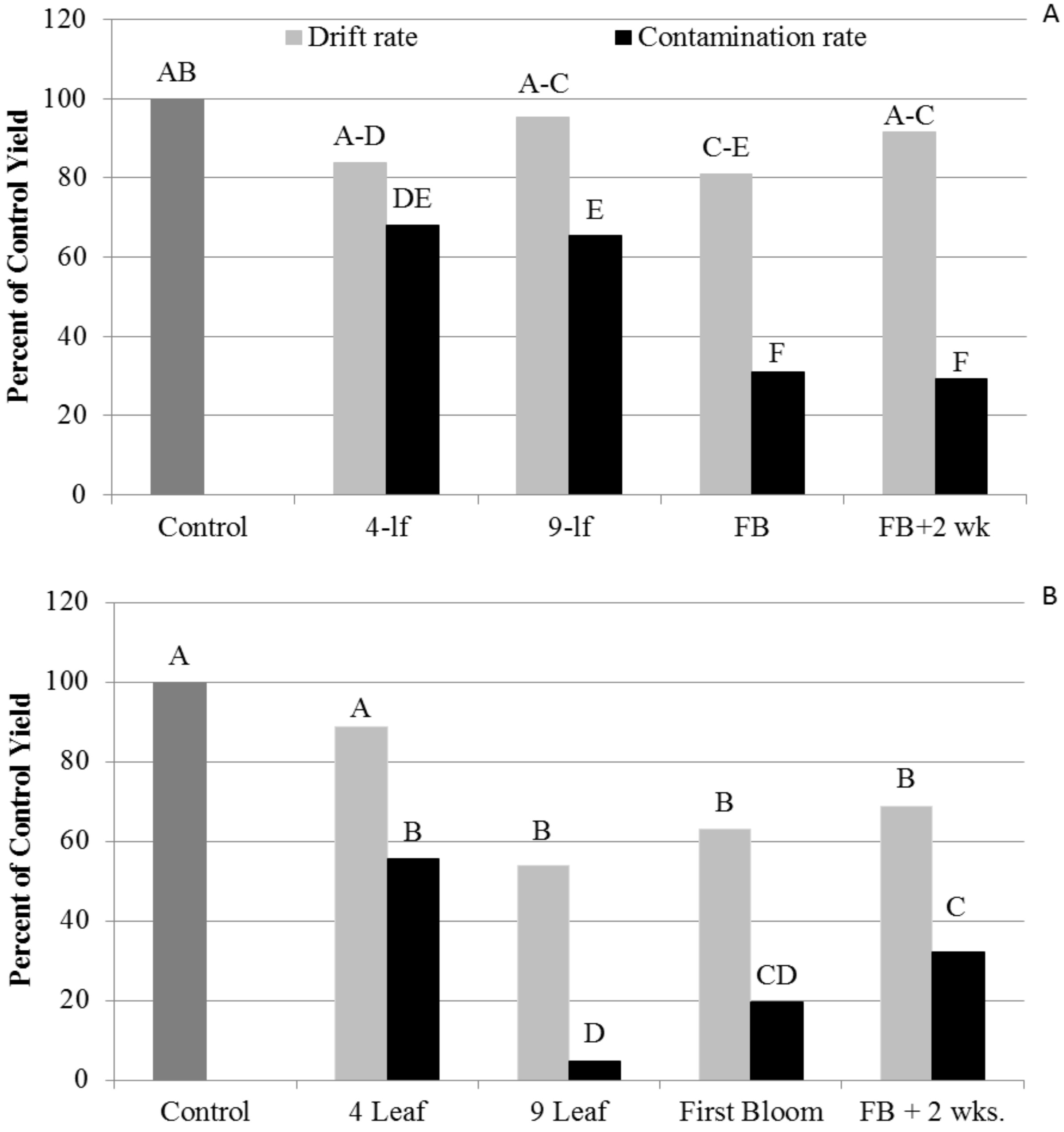

Figure 3.3. Percent of control yield of the drift rate and contamination rate of 2,4-D at the four leaf (4-lf), nine leaf (9-lf), first bloom (FB) and two weeks after first bloom (FB+2 wk) growth stages at Tifton (A) and Moultrie (B).

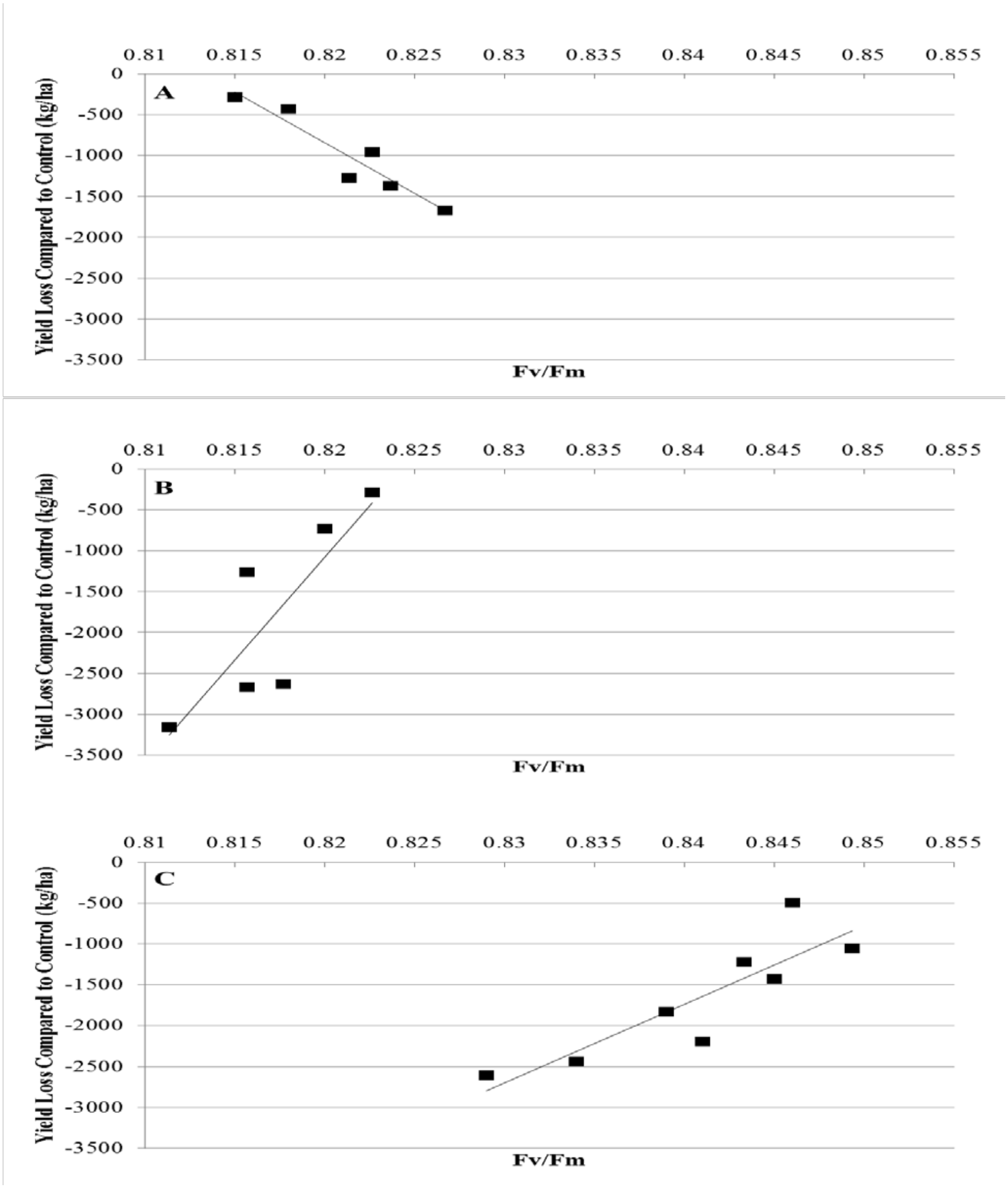

Figure 3.4. Regression results of the three instances of significant correlation between yield loss compared to the control treatment and F_v/F_m at the four leaf growth stage 14 DAA in Tifton (A), at the first bloom growth stage 6 DAA in Tifton (B), and at the two weeks after first bloom growth stage 22 DAA in Moultrie (C).

CHAPTER 4

IRRIGATION MANAGEMENT OF COTTON (*GOSSYPIUM HIRSUTUM*) IN CONSERVATION TILLAGE UTILIZING A HIGH BIOMASS RYE COVER CROP[1]

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[1] S.A. Byrd, G.D. Collins, R.M. Barentine, J.L. Snider, A.S. Culpepper, P.M. Roberts, W.M. Porter, and J.R. Whitaker. To be submitted to *Agronomy Journal*.

Abstract

Concerns surrounding agricultural water use have increased interest in investigating more efficient methods of crop production. Two experiments were conducted to determine potential water savings benefits of a conservation tillage system employing a high biomass rolled rye cover crop. The first experiment, conducted in Camilla, GA during 2013 and 2014 compared two tillage systems, conventional and conservation tillage with a rolled rye cover were evaluated under various irrigation levels (100, 75, and 50% of UGA recommendations, and nonirrigated/dryland) to determine the effect on soil moisture, crop growth and development, and yield of cotton. Also during 2014, a second experiment was conducted on-farm on large plots to compare the same two tillage systems in either an irrigated or a dryland environment. The rolled rye system increased soil moisture during 15 out of 86 measurements during 2013 and 2014 in Camilla. Several excessive rainfall event occurred in 2013, negatively impacting plant growth, development, and yield in the rye system, likely because of excessive soil moisture. During 2014 in Camilla where less than half of the rainfall occurred compared to the previous year, benefits in crop growth and development were observed with the rye but yield differences were not noted. The two additional large plot studies recorded similar results. No differences in plant growth were noted at the irrigated location throughout the season while decreased plant growth and development was noted in the dryland location, although no yield differences were noted at either large plot location. This study illustrates that the potential impact of cover crops from a water savings perspective is highly dependent on environmental conditions and is likely minimal under mild water deficit conditions, and perhaps detrimental when excessive rainfall occurs.

Introduction

Cotton is the most widely grown field crop in the state of Georgia and is a vital component of the state's economy as Georgia ranked second in the nation with 2.57 million bales produced in 2014 (USDA NASS, 2015). Increased regulations regarding water use at the federal (EPA, 2015) and state (GDNR – EPD, 2011; GDA, 2012) levels have heightened concerns and criticism over agricultural water use, particularly for irrigated producers who annually make up half of Georgia's cotton acreage. Regulations on water use, coupled with unpredictable rainfall patterns and episodic droughts occurring during the growing season make efficient use of water, whether rainfall or irrigation, a key for agricultural production.

While efficient water use is critical, avoiding the detrimental effects of inadequate water supply, or drought stress will also be a key for producers to maintain production that is economically sustainable. Yield loss due to water stress in cotton is well documented (Gerik et al., 1996; Pringle and Martin, 2003; Pettigrew, 2004; Whitaker et al., 2008; Gwathmey et al., 2011). Detrimental effects to fiber quality parameters such as fiber length (staple) and micronaire have been observed due to water deficit or stress in cotton (Grimes and Yamada, 1982, Pettigrew, 2004; Whitaker et al., 2008; Wen et al., 2013). One method that has the potential to mitigate both periods of low rainfall for nonirrigated (dryland) producers, and irrigation use under more stringent regulations, is a reduced or conservation tillage system with the utilization of a cover crop. Rye (*Secale cereale*) has become a popular cover crop choice in the Southeastern United States due to the ability to produce a high amount of biomass compared to other potential cover crop species (Bauer and Busscher, 1996; Daniel et al., 1999a; Sainju et al., 2005; Schomberg et al., 2006; Aulakh et al., 2012a). Rye has already gained additional recognition in Georgia as it has been shown to suppress Palmer amaranth (*Amaranthus palmeri*), the most problematic weed in the state, (Culpepper et al., 2006; Sosnoskie and Culpepper, 2012).

In this system, the rye is rolled and terminated with herbicides creating a mat of rye that covers the soil surface and prevents the germination of Palmer amaranth seeds by providing a barrier to sunlight reaching the seeds (Gallagher and Cardina, 1986a, 1986b; Cristaudo et al., 2007; Jha et al., 2010). A higher amount of biomass, or soil coverage provides greater weed control (Aulakh et al., 2012b; Norsworthy et al., 2011). Utilization of a rye cover crop has also been observed to reduce thrips (*Franklinielle fusca* [Hinds]) populations in cotton (Manley et al., 2002; Olson et al., 2006) with thrips suppression increasing with increased biomass (Olson et al., 2006).

Conservation tillage, including no-till and conservation tillage with a cover crop, has been observed in previous studies to increase soil moisture or soil water content compared to conventional tillage (Blevins et al., 1971; Gantzer and Blake, 1978; Mills et al., 1988; Dao, 1993; Daniel et al., 1999b), while also increasing infiltration and retention rates of water (Dao, 1993; Lascano et al., 1994; Bruce et al., 1995; Raper et al., 2000). However, the moisture saving benefits from a cover crop can be minimized or eliminated due to low cover crop biomass or heavy rainfall (Daniel et al., 1999b). A beneficial response to plant growth has been observed in cotton grown under conservation tillage with the utilization of a cover crop, including taller plants (Bauer and Busscher, 1996; Bauer et al., 2010) and a greater number of nodes (Wiatrak et al., 2005). A positive yield response of cotton under conservation tillage compared to conventional tillage has also been observed (Bordovsky et al., 1994; Bauer and Busscher, 1996; Raper et al., 2000; Schomberg et al., 2006; Wiatrak et al., 2006; Bauer et al., 2010) with rye providing a larger yield benefit than other cover crop species (Bauer and Busscher, 1996; Schomberg et al., 2006). Previous studies that investigated cotton fiber quality found no effect of conservation tillage or the utilization of cover crops on various fiber quality parameters (Smith and Varvil, 1982; Baker, 1987; Bauer and Busscher, 1996). While increasing plant growth, development, and yield are beneficial to cotton and all crops in general, the magnitude of the impact could be influenced by the maturity of cotton. Though the species is indeterminate by nature (Ray and Richmond, 1966), it is generally accepted that the maturity of cultivars varies, creating two primary categories; late and early maturing cultivars (Husman et al., 1996). These mainly refer to differences in fruiting habit, as early maturing cultivars typically set the majority of their fruit on a compact set of lower main stem nodes over a shorter period of time than late maturing cultivars which have a greater vertical distribution of fruit that is set over a longer period of time compared to early maturing cultivars (Husman et al., 1996; Bednarz and Nichols, 2005). Early season cultivars are more prone to short-term periods of water deficit, due to a shorter boll development period compared to late season cultivars which can mitigate these short term deficit periods with an extended fruiting period (Rosenow et al., 1983; Husman et al., 1996; Snowden et al., 2013). A conservation tillage system that utilized a cover crop could presumably provide additional benefits for weed and thrips suppression including increased soil moisture allowing for better crop performance, particularly during periods of unfavorable environmental conditions.

The objectives of the current study were to quantify the effect of cotton grown in the high rye biomass conservation tillage system that has been successful in Palmer amaranth suppression compared to a conventional tillage system. Two primary scenarios were the focus of the study; large fields with large irrigation systems which can have difficulty applying the recommended rates of irrigation during periods of low rainfall, and dryland situations where producers rely on rainfall as their sole source of water. In both scenarios, the ability of the high rye biomass system to retain moisture in the soil profile could mitigate the strain placed on crops during dry periods, assist in avoiding periods of drought stress, and allow for more efficient irrigation or a reduction in the amount of irrigation required.

Materials and Methods

Methods Specific to Stripling. Research was conducted at the University of Georgia's C.M. Stripling Irrigation Research Park in Camilla, GA (31°16'46" N, 84°17'48" W) during the 2013 (Camilla 2013) and 2014 (Camilla 2014) cotton season, on separate sections of the same field. The 2013 crop followed cotton, while the 2014 crop followed peanuts. The soil type at this location is a Lucy loamy sand (USDA NRCS 2015a) classified as a loamy, kaolinitic, thermic Arenic Kandiudult and being very deep, well drained, and moderately permeable (USDA NRCS 2015b). A randomized complete block designed was utilized with a factorial arrangement of treatments.

Wrens Abruzzi rye was drilled at a 101 kg ha^{-1} seeding rate with a Great Plains 3P606NT grain drill (Great Plains Ag, Salina, KS) each November prior to the following cotton season, only in plots that would serve as conservation tilled plots the following cotton season. Two weeks after planting, 22 kg ha^{-1} of nitrogen was applied to the plots containing rye. The grain drill had been modified to plant rye seed in rows 15 cm apart except where each cotton row would be planted where 30.5 cm wide spacing was placed. The goal of the rye plant spacing was to ensure the rye cover would not impede the emergence of cotton. The rye was rolled with a 2.4 m wide roller crimper (I & J Manufacturing, Gap, PA) and sprayed with a burndown herbicide consisting of 1.61 L ha^{-1} of glyphosate and 0.15 L ha^{-1} of flumioxazin approximately two weeks prior to cotton planting. Rye biomass was measured by sampling a 30.5 cm^{-2} area prior to the rye being rolled. The aboveground biomass was harvested, then dried and weighed to quantify the dry biomass present.

Two tillage treatments were included in the study, conventional tillage and the conservation tillage rolled rye cover system. Plots in conventional tillage contained no rye cover and were planted with the same strip tillage implement utilized for planting the conservation tillage plots. A Monosem STD 540 (Monosem Inc. Edwardsville, KS) two row vacuum planter mounted behind a KMC strip tillage implement (Kelly Manufacturing Co. Tifton, GA) to facilitate planting operations into rye cover was utilized for cotton planting in both the conservation and conventional tillage plots. The strip tillage implement included a cutting blade, ripper shank, row cleaners, and planting disc for each row. Because the strip till implement was utilized for planting both conventional and rye treatments, only an additional pass of a rototiller was performed on conventionally tilled plots prior to planting. Four irrigation treatments were also utilized in the study. Irrigation recommendations for high yielding cotton in Georgia are provided by the University of Georgia checkbook (Table 4.1). The irrigation treatments in this study consisted of the levels recommended by the UGA checkbook schedule (100% checkbook), 75% of the UGA checkbook recommended levels (75% checkbook), 50% of the UGA checkbook recommended levels (50% checkbook) and a non-irrigated treatment (dryland). Irrigation applications were made to supplement rainfall so that each treatment reached its respective recommended target of total water for each week. Irrigation was split into two applications per week to account for rainfall and to prevent excessive runoff from higher applications. Plots consisted of two 91.5 cm rows 13.7 m in length. All crop management practices, including an additional 22 kg ha^{-1} of nitrogen applied to cotton grown in the conservation tillage plots approximately two weeks after planting, followed extension recommendations (Collins et al., 2015). The early maturing cultivar ‘FiberMax 1944 GLB2’ (FM 1944, Bayer CropScience, Research Triangle Park, NC) and the later maturing cultivar ‘PhytoGen 499 WRF’ (PHY 499, Dow AgroSciences, Indianapolis, IN) were planted in both years of the study. A randomized complete block design was utilized, with each cultivar by irrigation by tillage treatment combination replicated three times, and each replicate containing every possible treatment combination. Cotton was planted on 29 April in 2013 and on 9 May in 2014 at a rate of 11.5 seeds m^{-1}.

Methods Common to Large On-Farm Trials. Two additional large experimental sites were evaluated in 2014. An irrigated site at the Sunbelt Agricultural Exposition in Moultrie, GA (31°08’25” N, 83°43’08” W) was included and contained four replications of conventionally tilled and conservation tilled treatments, alternated across the field. The conservation tilled plots included a rolled rye cover crop planted and managed following the same methods as previously described at Camilla. Irrigation was uniform over all plots and supplemented rainfall to reach 100% UGA Checkbook levels each week. The soil type at this location is a Leefield loamy sand (USDA NRCS 2015a) classified as a loamy, siliceous, subactive, thermic Arenic Plinthaquic Paleudult and being very deep, somewhat poorly drained, and moderately slowly to slowly permeable (USDA NRCS 2015b). Plots in the Moultrie location consisted of four rows of cotton spaced 91.5 cm apart with plot lengths varying from 110 to 218 m. The cultivar FM 1944 was planted at the Moultrie location on 6 May 2014 at a rate of 11.5 seeds m^{-1}. An on-farm trial was included during 2014 in Vienna, GA (32°08’22” N, 83°47’24” W). The Vienna site was nonirrigated and included four replications of conventionally tilled and conservation tilled plots with a rolled rye cover. The tillage treatments were alternated across the field, with each rep containing a conventional and conservation tilled plot. At this location, Elbon rye was drilled in

19 cm rows at 90 kg ha^{-1} seeding rate with John Deere 1590 (John Deere, Moline, IL) with no gap left in the rye for cotton planting. The soil type at this location is a Dothan loamy sand (USDA NRCS 2015a) classified as a fine-loamy, kaolinitic, thermic Plinthic Kandiudult and being very deep, well drained, and moderately slowly to slowly permeable (USDA NRCS 2015b). The cultivar PHY 499 was planted with a John Deere 1700 planter (John Deere, Moline, IL) which included a strip tillage implement ahead of the planter on 12 May, 2014 at a seeding rate of 10 seeds m^{-1}. Plots consisted of eight rows of cotton spaced 96.5 cm apart with plots varying from 234 to 338 m in length. Cotton was planted in the previous year in both the Moultrie and Vienna locations. All other crop management practices at Moultrie and Vienna followed extension recommendations (Collins et al., 2015).

Methods Common to All Locations. At all locations, soil moisture was quantified once per week at 10 and 20 cm depths with Spectrum FieldScout TDR 300 soil moisture meters (Spectrum Technologies Inc. Aurora, IL). Three readings of volumetric soil water content at each depth were taken within the row in each plot and averaged to give one value per plot each week. In both years at the Camilla locations, soil moisture readings were initiated one to two weeks before the rye was terminated and rolled so that the effect of the cover crop on soil moisture prior to planting could be quantified. To determine the impact of the tillage and irrigation treatments, plant growth and reproductive development measurements were taken throughout the season. Measurements included plant height at the eight leaf (8-lf) stage, which is typically the start of the squaring stage, during the first week of bloom, or early bloom (EB), and at two, four, and six weeks after EB (EB+2 wk, EB+4 wk, and EB+6 wk, respectively). Plant heights at the 8-lf stage were measured prior to the beginning of the irrigation treatment, which was initiated at this growth stage. Nodes above the uppermost first position white flower (NAWF) has become a standard measurement for determining physiological maturity, or cutout, of cotton (Oosterhuis et al., 1992; Bourland et al., 2001), with any bolls developing beyond NAWF = 5 contributing very little to the final yield (Oosterhuis et al., 1992). NAWF measurements were taken at EB, EB+2 wk, EB+4 wk, and EB+6 wk growth stages. Plant growth measurements at the EB+2wk growth stage were omitted from the Camilla 2014 location. Plant mapping was performed near the end of the season, prior to defoliation, on seven plants per plot, with measurements including plant height, total nodes, and total bolls.

Defoliation applications were made when the majority of plants had reached the stage of four nodes above the uppermost first position cracked boll (Collins et al., 2015). Harvest occurred on 9 October 2013, 2 October 2014, 6 October 2014, and 20 October 2014 for the Camilla 2013, Moultrie, Camilla 2014, and Vienna locations, respectively. Plots at the Camilla and Moultrie locations were harvested with a John Deere 9930 (John Deere, Moline, IL) two-row spindle picker with a bagging attachment installed at Camilla. Seedcotton weights at Camilla were taken after harvest using Intercomp CS750 digital scale (Intercomp Company, Medina, MN). At Moultrie, four rows of each plot were harvested and dumped into a boll buggy which had load cells installed to quantify the weight of seedcotton inside the buggy. Plots at the Vienna location were harvested with a John Deere 9970 (John Deere, Moline, IL) four row picker and weighed on site with the same boll buggy utilized at the Moultrie location for seedcotton weight determination. Seedcotton samples were collected and ginned at the University of Georgia Micro Gin in Tifton, Georgia (Li et al., 2011) for determination of lint percentage and lint yield. For the Moultrie and Vienna locations, because the same cultivar was used within each trial, only one sample was analyzed for lint percentage and this value was applied to all seedcotton samples to determine lint yield. For cotton harvested from Camilla, approximately 230 g of lint from each plot were sent to the USDA Classing Office in Macon, GA after ginning for both classing and high volume instrumentation (HVI) measurements of fiber quality.

Statistical analysis was performed using SAS 9.4 software (SAS Institute Inc., Cary, NC) with the macro pdmix 800 (Saxton, 1998). Tillage and cultivar served as the fixed effects, and irrigation as the random effect. Treatment means were separated by Fisher's Protected LSD at an alpha level of ≤ 0.05. When all data was pooled, the main effect of location was significant for both seedcotton and lint yields, thus locations were analyzed independently.

Results and Discussion

Climate. Weekly rainfall and irrigation amounts (at the 100% checkbook level) for the irrigation treatment period, as well as early and late season total amounts are presented in Table 4.2 for all four locations. The total amount of water received (irrigation and rainfall) at the Camilla 2013 location was 95.9 cm in the 100% checkbook, 91 cm in the 75% checkbook, 86.3 cm in the 50% checkbook, and 76.8 cm in the dryland treatments. In Camilla 2014, the total amount of water received (irrigation and rainfall) was 66.4 cm in the 100% checkbook, 57.7 cm in the 75% checkbook, 49.2 cm in the 50% checkbook, and 36.2 cm in the dryland treatments. The dryland treatment at Camilla received 2.3 cm and 1.8 cm of irrigation with fertilizer applications and for herbicide activation early in the season in 2013 and 2014, respectively. Total water received (irrigation and rainfall) exceeded targeted amounts multiple weeks in both years at Camilla. This is primarily a result of one of two scenarios; irrigations being made during the first half of the week followed by heavy precipitation events later in the week, or precipitation alone exceeded the targeted amount.

Rye Biomass. The conservation tillage treatment resulted in an average of 12,015 kg ha^{-1} of dry rye biomass at the Camilla 2013 location, 17,165 kg ha^{-1} of dry rye biomass at Camilla 2014, 6,077 kg ha^{-1} at Moultrie, and 11,046 kg ha^{-1} at Vienna.

Soil Moisture. Measurements of soil moisture prior to the rye being rolled and killed showed increased soil moisture in the conventional tillage plots at one out of two weeks at the 10 cm depth and in both weeks at the 20 cm depth in Camilla 2013 (data not shown). This decline in soil moisture under the conservation tillage is expected with water uptake by the rye cover. In the two readings following the termination and rolling of the rye before planting, soil moisture was increased in both weeks at both depths in the conservation tillage plots. This result is likely in response to rainfall occurring after killing the rye that replenished the soil moisture and the dead cover crop then reducing water loss. Throughout the season in the dryland treatment, measurements of soil moisture in the rye system showed increased soil moisture at the 10 cm depth in four out of 13 weeks and in five out of 13 weeks at the 20 cm depth. Soil moisture was taken in eight weeks after the irrigation treatments were initiated. During this period there was only one instance of a significant increase in soil moisture in the conservation tillage treatment, which occurred at the 20 cm depth in the 50% checkbook treatment. Across the 2013 season there were three instances in which higher soil moisture was measured in the conventional tillage treatment, at 20 cm in the dryland treatment, and at both depths in the 75% checkbook treatment. In the Camilla 2014 location, the only differences in soil moisture were observed in the 75% checkbook irrigation treatment, where in four out of 12 measurements at 10 cm, higher soil moisture was present in the conservation tillage treatment, and in one out of 12 measurements soil moisture was higher at 20 cm in the conservation tillage treatment (data not shown). In Moultrie, soil moisture was higher in the conservation tillage treatment at both 10 and 20 cm in two out of 11 measurements (data not shown). The greatest number of significant differences in soil moisture was observed at the Vienna location, where lower rainfall amounts were recorded; soil moisture in the conservation tillage treatment was greater at the 10 cm depth in 10 out of 17 measurements, while soil was greater at 20 cm in three out of 17 measurements (data not shown).

Increased soil moisture is one of the key reasons the high biomass rye system is thought to have potential for increasing irrigation efficiency and mitigating drought stress, however, in the current study the amount of instances in which this system increased soil moisture compared to conventional tillage were relatively small. The differences present in soil moisture prior to planting between Camilla 2013 and 2014 are likely due to rainfall events leading up to planting. In the approximately two weeks leading up to planting in 2013, 1 cm of rainfall was received, while over 7.5 cm of rain fell during this same time period during 2014. The increase in soil moisture with conservation tillage illustrates the benefits of conservation tillage during periods of dry weather, allowing for a moist seed bed to be present at time of cotton planting. Frequent rainfall received in 2014 prior to planting likely minimized any additional moisture retention benefit from the rolled rye cover. The relatively small number (10 out of 37 across all treatments) of instances in 2013 of increased soil moistures in the conservation tillage treatment is likely a result of frequent, heavy rainfall events occurring throughout the season. Previous studies have reported increased rainfall infiltration rates when comparing conservation tillage, including the utilization of a cover crop, to conventional tillage (Raper et al., 2000; Bruce et al., 1995; Dao, 1993), which may be the primary cause of the few instances in which soil moisture was greater in the conventional tillage treatment. These situations may reflect saturated soil conditions in conventionally tilled plots in the shallow depths at which soil moisture was measured in the current study. It is unclear why few instances of increased soil moisture during the growing season were observed in the Camilla 2014 location, particularly that all differences were confined to the 75% checkbook treatment. The low number of instances of soil moisture differences observed between tillage treatments at the Moultrie location (4 out of 22 across both depths) is likely due to differences in soil characteristics, particularly the drainage classification, and the lower amount of rye biomass at Moultrie compared to the other locations. The greatest success for increasing soil moisture under conservation tillage was observed at the Vienna location. Several factors may have influenced this, including the amount of biomass, approximately 5,000 more kg ha^{-1} than the other large plot trial in Moultrie, and the lack of irrigation, similar to soil moisture results from the dryland treatment at Camilla 2013. The effects of cover crops on soil moisture have been observed to be minimized in the past due to a low amount of cover crop biomass and heavy rainfall during the season (Daniel et al., 1999b) which may explain the lack of numerous instances of significant differences in the current study.

Plant Height. There were no significant interactions present between any of the main effects for plant height or NAWF in either year in Camilla, as well as no effect of tillage treatment on any plant growth measurements at the Moultrie location. The main effects of cultivar, tillage, and irrigation were significant at all five measurement dates for plant height at Camilla 2013 (Table 4.3). The cultivar PHY 499 was taller at all measurement dates, while cotton grown under conservation tillage was shorter at all dates (Table 4.4). At the beginning of squaring (8-lf) few differences were noted between irrigation schedules with cotton height ranging from 28 to 30 cm. This is expected as irrigation treatments had not initiated at this point. By EB, the benefits of irrigation were noted with cotton ranging from 52 cm in dryland to 59 cm with the 100% checkbook. By late-season, cotton height was similar when comparing 75 and 100% checkbook, taller than cotton in 50% checkbook, with the shortest cotton in dryland treatment. At Camilla in 2014, the effect of cultivar on plant height was significant at three of five evaluation dates while tillage was significant in two out of the four dates. Similar to 2013, PHY 499 was taller than FM 1944 after the 8-lf stage. Conventional tillage resulted in plants 4.9 cm taller compared to conservation tillage at the 8-lf measurement, while at EB+4 wk plants under conservation tillage were 6.8 cm taller than those in conventional. Similar to 2013, no differences between 75 and 100% checkbook were noted. Additionally, cotton receiving at least 75% checkbook amounts was taller than cotton that received 50% checkbook and dryland at EB+4 wk and EB+6 wk. Over the entire season, tillage had no effect on plant height at the Moultrie location. With the exception of the 8-lf measurement, conventional tillage resulted in taller plants throughout the season at Vienna, with differences ranging from 7.4 – 21.9 cm throughout the bloom period.

When significant differences were present in plant height due to cultivar, this is an illustration in the differentiation in growth habits between early and late maturing cultivars. Early maturing cultivars develop bolls over a more compact set of nodes, leading to a higher boll load earlier in the season than late maturing cultivars which have a greater vertical distribution of bolls over a larger range of nodes (Bednarz and Nichols, 2005; Husman et al., 1996). This increased early season boll load leads to decreased vegetative growth resulting in shorter plants in early maturing cultivars. At both the Camilla 2013 and Vienna locations, shorter plants resulted from the conservation tillage treatment, while at Camilla 2014 the conservation tillage resulted in taller plants compared to conventional tillage. Thus, at two out of three locations where tillage had a significant effect on plant height, the results of the present study contrast results of increased plant heights with conservation or reduced tillage observed in previous studies (Bauer et al., 2010; Bauer and Busscher, 1996). These findings are also in contrast to the soil moisture results at Vienna, where soil moisture was increased in the majority of readings at the 10 cm depth. However, Bauer et al. (2010) observed increased plant height during a drought period, conditions that were not present in any of the four locations in the current study. At Camilla in 2013, weekly rainfall totals in excess of the 100% checkbook recommendations fell during weeks one, two, three, and five of the bloom period, which is likely the reason for shorter plants at EB+6 wk in the 100% checkbook treatment compared to the 75% checkbook treatment. Increases in irrigation generally resulted in increases in plant height throughout both seasons at Camilla, although there was often no difference in plant height between the 75 and 100% checkbook treatments.

NAWF. Tillage and irrigation were significant at all growth stages for NAWF in Camilla 2013, while cultivar was significant at the initial EB measurement (Table 4.5). FM 1944 resulted in greater NAWF at only the EB measurement, while NAWF were greater in conventional tillage during all measurement dates except EB+2 wk (Table 4.6). Conventional tillage increased NAWF by 1.7, 0.6, and 2.1 nodes at EB, EB+4 wk, and EB+6 wk, respectively, which would be expected with taller cotton (Table 4.5) (Pettigrew, 2004; Bauer et al., 2010). All irrigated treatments resulted in increased NAWF compared to dryland during the first two measurements with few differences noted later in the season as the crop matured. Cultivar had a significant effect at one measurement in Camilla during 2014, as PHY 499 had only 0.7 more nodes than FM 1944 at only the EB+4 wk date. Tillage was again significant at all measurement dates. In contrast to 2013, the rye system had 0.8 to 0.9 more nodes than the conventional system. Differences were likely in response to rainfall differences noted during the two years (Table 4.2). During 2014 with less rainfall, taller plants later were present in the rye system (Table 4.4) resulting in more NAWF as expected. Also in contrast to 2013, irrigation scheduling had a much greater impact on node development, with more NAWF noted with higher rates in the 75 and 100% checkbook treatments. Similar to height measurements, tillage had no effect on NAWF at Moultrie. At Vienna, conventional tillage resulted in almost one more NAWF at EB+2 and EB+4 wk.

Reflective of plant height results, greater NAWF were observed as a result of the conventional tillage treatment at Camilla 2013 and Vienna 2014, while conservation tillage increased NAWF in Camilla 2014, when significant. Excess water likely also led to a decrease in NAWF in the 100% checkbook compared to the 75% checkbook at EB+4 and EB+6 wk measurements. Though it appears there are no previous studies on the effect of excess water on cotton in the southeast, likely because these events are rare particularly on the low water holding capacity soils prevalent in the region, prolonged saturated soil conditions can have detrimental impacts on crop growth and performance (Kozlowski, 1984). In 2014, when less than half the 2013 rainfall was received, the 100% checkbook treatment consistently resulted in NAWF no different from the 75% checkbook (EB and EB+4 wk) or greater than all other treatments (EB+6 wk) illustrating the value of supplemental irrigation when crop water requirements are not met by rainfall alone and agreeing with previous studies (Pettigrew, 2004; Whitaker et al., 2008).

Plant Mapping. The main effects of cultivar, tillage, and irrigation were significant for end of season height measurements at Camilla 2013 (Table 4.7). The cultivar PHY 499 resulted in taller plants, while plants grown in conventionally tilled plots were 8.5 cm taller than those in conservation tillage, reflecting the height measurements recorded earlier in the season (Table 4.8). There was no difference in plant height across any of the treatments that received irrigation; however, the heights across all three were greater than the dryland treatment. Total nodes but not total bolls were influenced by cultivar, with 1.5 more nodes in FM 1944 compared to PHY 499. For tillage, total nodes and total bolls 20 and 38% less with the rye system compared to conventional tillage, respectively. In Camilla during 2014, no differences in total nodes were detected but approximately 10 more bolls were noted with PHY 499 as compared to FM 1944. Tillage comparisons noted no differences in regards to total bolls, but did note 1.1 more nodes per plant in the rye system as expected with taller plants (Table 4.4). The effect of irrigation on end of season plant heights reflected the results of height measurements taken during the season, with taller plants in the 100% and 75% checkbook treatments, while the 50% checkbook treatment resulted in taller plants than dryland. Irrigation also had a significant effect on total nodes, with total nodes declining with each reduction in irrigation. The number of total bolls was highest under the 100% and 75% checkbook irrigation treatments (110.1 and 98 bolls, respectively), followed by the 50% checkbook which had a significantly higher number of bolls (71.9) than the dryland treatment (52.4). In Moultrie, tillage had no effect on total nodes or bolls, while in Vienna there were 0.6 more total nodes in the conventional tillage system.

The number of total nodes followed the same pattern as plant heights in regards to tillage. Increases in plant height and total nodes with increasing water supply are in agreement with previous research (Pettigrew, 2004; Bauer et al., 2010).

Yield. While cultivar had no effect on seedcotton yield at the Camilla 2013 location, differences in both lint percentage and lint yield were observed (Table 4.9). PHY 499 had greater values in both parameters than FM 1944 (Table 4.10). This is likely due to FM 1944 being a larger seeded cultivar than PHY 499, producing a lesser proportion of lint in the harvested seedcotton resulting in a greater amount of lint yield (Miller and Rawlings, 1967; Culp and Harrell, 1975). Tillage influenced all three yield parameters, and though lint percentage was greater by 1.5% in the conservation tillage treatment, conventional tillage increased seedcotton yield by 812 kg ha^{-1} and lint yield by 261 kg ha^{-1}. This reflects the shorter plants, more rapid decline in NAWF, and total number of bolls observed through the course of the season. The dryland irrigation treatment resulted in lint percentage values lower than the 50 and 100% checkbook irrigation treatments. Cultivar had a significant effect on seedcotton yield, lint percentage, and lint yield at the Camilla 2014 location, with values of all three parameters higher in PHY 499. In contrast to the Camilla 2013 results, conventional tillage led to a 0.5 increase in lint percentage, but there was no tillage effect on either seedcotton or lint yield. Irrigation had a significant effect on all yield parameters, with the 75% checkbook treatment resulting in the highest values in all three, though no different from the 100% checkbook in regard to lint percentage and lint yield. The 75% checkbook treatment produced 234 kg ha^{-1} more lint yield than the 50% checkbook treatment and 758 kg ha^{-1} more than dryland. For Moultrie and Vienna, tillage did not influence yield.

It is not surprising that PHY 499 outperformed FM 1944 in terms of yield parameters, as these results agree with the University of Georgia on-farm variety trials (Collins and Whitaker, 2014). Lack of significant interactions between cultivar and tillage or irrigation suggest that differences observed between cultivars, namely plant growth and development and fiber quality, are characteristics related to differences in the cultivars and are not due to the tillage and irrigation treatments. The decline in the number of total bolls in the conservation compared to the conventional tillage treatment in Camilla 2013 is reflected in the reduction of seedcotton and lint yield, as Krieg (2000) reported that the number of bolls is responsible for 85% of yield variability in cotton. This may also explain why irrigation had no effect on yield, as it similarly did not influence the number of bolls. Irrigation had an impact on yield at Camilla 2014, likely due to a lower amount of total rain at the Camilla location in 2014 (36.2 cm) compared to 2013 (76.8 cm) again emphasizing the value of supplemental irrigation. Another key finding from this year was the lack of difference in lint yield between 100 and 75% checkbook treatments, which mirrors the observations of plant height and NAWF taken throughout the season. This provides evidence that perhaps slight reductions could be made in irrigation recommendations, as the 75% checkbook treatment applied 8.7 fewer cm of irrigation than the 100% checkbook treatment no impact on lint yield, and an increase in seedcotton yield. The yield patterns also reflected the total boll numbers as affected by irrigation, which agrees with the findings of previous research (Turner et al., 1986; Bauer et al., 2010). The increase in lint percentage at Camilla in 2013 in the conservation tillage treatment is similar to the findings of Bauer and Busscher (1996) who reported a 1% increase in lint percentage comparing conservation to conventional tillage. All irrigation treatments resulted in higher lint percentage than the dryland treatment in both years of the study at the Camilla location. Few instances of significant differences in lint percentage have been observed in previous comparisons of irrigated and dryland cotton production. Greater lint percentage has been observed from either increases or reductions in water supply, though the majority of the time there are no significant differences (Pettigrew, 2004; Whitaker et al., 2008; Gwathmey et al. 2011).

Fiber Quality. The only instances of significant interactions between the main effects occurred in fiber quality parameters (Table 4.11). At the Camilla 2013 location, a significant effect of cultivar by tillage resulted in increased micronaire in PHY 499 under conservation tillage compared to the three other cultivar by tillage combinations (Fig. 4.1A). A significant cultivar by irrigation effect was present for fiber strength, with dryland FM 1944 resulting in greater fiber strength than all other cultivar by irrigation combinations except PHY 499 at 50% checkbook (Fig. 4.1B). A significant tillage by irrigation interaction was also present for fiber strength, with the conventionally tilled dryland treatment resulting in higher fiber strength than all other tillage by irrigation combination with the exception of conventional tillage at 75 and 50% checkbook and the dryland conservation tillage treatment (Fig. 4.1C). The three way interaction of cultivar by tillage by irrigation was significant for fiber length uniformity, with lower uniformity typically resulting from FM 1944 under both tillage treatments and all irrigation levels with the exception of conventional tillage 75% checkbook treatment, and PHY 499 under dryland conventional tillage (Fig. 4.2). At the Camilla 2014 location, there was a significant tillage by irrigation effect on micronaire, with values higher in the conventionally tilled 100% checkbook treatment compared to all other tillage by irrigation combinations (Fig. 4.1D). The main effects of cultivar, tillage, and irrigation were significant for staple in both years at Camilla (Table 4.11). All treatment means fell within a range 2/32^{nds} of an inch (the official unit of measure for staple; USDA AMS, 1995) with the largest difference resulting from cultivar at the Camilla 2014 location (Table 4.12). FM 1944 resulted in micronaire values that fell within the premium range compared to the base range of values of micronaire (USDA AMS, 1995) found with PHY 499 at Camilla 2014. Cultivar was also significant for fiber uniformity at Camilla 2014, with increased uniformity resulting from PHY 499. Categories for ranges of micronaire, fiber strength, and fiber uniformity are used when classifying cotton lint (USDA AMS, 1995). In the present study, the only parameter than had a difference in treatments that led to a realistic difference in fiber quality was micronaire, as all values for strength fell into the “very strong” category, while all fiber length uniformity value were classified as “high” (USDA AMS, 1995). The PHY 499 rye treatment at Camilla 2013 resulted in micronaire values (50) that were in the discount range, or greater than optimal micronaire values, with micronaire values of all other treatments falling into the base range. At Camilla 2014, values of micronaire in the 50% checkbook and dryland rye treatments fell into the premium range (37-42) with all other treatments resulting in base range values (35-36; 43-49) (USDA AMS 1995).

The only parameter that showed a realistic significant difference in terms of fiber quality classification due to tillage was micronaire. The conservation tillage treatment in 2013 resulted in micronaire falling into the discount range, while conventional tillage micronaire fell into the base range of values (USDA AMS 1995). In 2014, the conservation tillage treatment resulted in micronaire in the premium range, while base range micronaire again resulted from conventional tillage. The minimal amount of realistic differences for fiber quality parameters among tillage treatments agrees with findings of previous studies (Smith and Varvil, 1982; Baker, 1987; Bauer and Busscher, 1996). While previous studies have documented detrimental effects of water deficit or drought conditions on the fiber quality parameters quantified in the present study (Grimes and Yamada, 1982, Pettigrew, 2004; Whitaker et al., 2008; Wen et al., 2013), due to the amount of rainfall received in both years at Camilla water deficit conditions severe enough to impact fiber quality did not exist.

Summary. While utilization of the high rye biomass conservation tillage system did result in numerous instances of increased soil moisture, across all four locations and both depths of the study these constituted only 8% of soil moisture readings. While scarce, benefits to plant growth and development were observed, particularly in Camilla 2014, with no detrimental effect of yield was observed, with the exception of the Camilla 2013 site at which excessive rainfall amounts were received throughout the season. In regards to irrigation, it was evident that when excessive rainfall occurred, such as was the case at Camilla in 2013, there is little benefit from irrigation, as no supplementation is necessary. Perhaps one of the most critical findings from the Camilla 2014 site was the minimal difference in plant growth found between 75% and 100% checkbook irrigation levels, resulting in no difference in lint yield. Achieving this type of yield accompanied water savings has serious potential for additional work in the southeast regarding irrigation recommendations. However, further research would need to be done to transition findings from one year of work into recommendations for crop management.

There are, however, additional methods that could be implemented to further investigate and understand the potential water savings benefits of the high biomass rye conservation tillage system. A more intensive method of soil moisture determination, such as using sensors to log soil moisture at more frequent intervals, rather than weekly, point-in-time measurements could improve comprehension of the water savings throughout the season. If this type of soil moisture monitoring had been in place, a more comprehensive understanding of the impact of the excessive rainfall received in 2013 could have been gained and related to the effects on plant growth and yield, as weekly measurements showed little to no difference in soil moisture between tillage treatments during this time period. Another desirable situation from a cropping systems perspective would be the continuation of this type of high biomass study in the same location over a series of consecutive growing seasons. At all four locations of this study, due to rotational patterns, the locations had not previously been under conservation tillage. Establishing a location for high biomass rye conservation tillage research for cotton, in which comparisons to conventional tillage could be made over numerous, continuous site years, may exhibit benefits from the development and continual implementation of this system over a long period of time.

Given that instances of improved soil moisture and crop growth and development, though few, were present in the current study, this high biomass rye system does show some potential for benefits regarding water savings and crop performance. The already established benefits of suppression of Palmer amaranth and thrips, two of the most detrimental pests to cotton production in the Southeast, will continue to make this system an attractive option for cotton producers seeking more efficient overall production practices.

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Table 4.1. Cotton Irrigation Schedule Suggested for High Yields

Crop Growth Stage	cm per week	cm per split application†
Beginning of squaring to 1st bloom	2.5	1.25
Week beginning at 1st bloom	2.5	1.25
2nd week of bloom	3.8	1.9
3rd week of bloom	5.1	2.55
4th week of bloom	5.1	2.55
5th week of bloom	3.8	1.9
6th week of bloom	3.8	1.9
7th week of bloom and beyond	2.5	1.25

†cm of irrigation per application if split into two applications per week

Taken from the 2015 Georgia Cotton Production Guide (Collins et al., 2015).

Table 4.2. Rainfall and irrigation (100% checkbook) received (cm) at each location.

Growth Stage	Source	Camilla 2013	Camilla 2014	Moultrie 2014	Vienna 2014
Planting – 1st square	Rainfall	14.9	16.5	26.6	8.8
Squaring Wk. 1	Irrigation	0.0	1.9	1.2	
	Rainfall	5.8	1.4	1.6	1.7
Squaring Wk. 2	Irrigation	1.8	1.3	2.2	
	Rainfall	3.4	4.5	0.4	3.9
Squaring Wk. 3	Irrigation	2.5	2.5	0.3	
	Rainfall	0.0	0.0	2.2	0.5
Bloom Wk. 1	Irrigation	1.3	2.5	0.0	
	Rainfall	5.6	0.1	5.7	3.8
Bloom Wk. 2	Irrigation	0.0	2.5	0.8	
	Rainfall	14.3	1.7	3.0	1.9
Bloom Wk. 3	Irrigation	1.8	4.1	4.1	
	Rainfall	5.9	1.2	1.0	1.0
Bloom Wk. 4	Irrigation	4.7	5.1	5.1	
	Rainfall	1.4	0.4	0.0	0.0
Bloom Wk. 5	Irrigation	0.0	3.8	2.1	
	Rainfall	5.4	0.0	1.7	1.0
Bloom Wk. 6	Irrigation	3.8	3.3	3.7	
	Rainfall	0.5	0.5	0.1	1.8
Bloom Wk. 7	Irrigation	1.9	0.8	0.0	
	Rainfall	0.6	1.8	2.9	0.0
Bloom Wk. 8	Irrigation	1.3	2.3	0.7	
	Rainfall	16.4	2.1	1.8	1.0
End of Bloom - Harvest	Rainfall	6.6	12.6	24.4	3.6
Total		99.9	72.9	91.6	29.0

Table 4.3. Anova results (F-values) of height measurements from Camilla 2013 and 2014, Moultrie 2014, and Vienna 2014 locations. Factors include cultivar, tillage, irrigation, two way interactions of cultivar by tillage, cultivar by irrigation, and tillage by irrigation, and the three way interaction of cultivar by tillage by irrigation.

Factors	df	8-lf	EB	EB+2 wk	EB+4 wk	EB+6 wk
Camilla 2013						
Cultivar‡	1	13.42***	39.49***	33.41***	36.67***	26.82***
Tillage§	1	16.35***	37.77***	7.22*	34.99***	71.44***
Irrigation¶	3	3.21*	10.35***	24.74***	11.52***	16.98***
Cultivar*Tillage	1	1.01	0.06	0.24	1.39	0.99
Cultivar*Irrigation	3	1.40	0.56	0.58	0.15	0.32
Tillage*Irrigation	3	2.41	0.34	0.46	0.11	1.77
Cultivar*Tillage*Irrigation	3	0.29	0.66	0.66	0.12	0.05
Camilla 2014						
Cultivar	1	1.14	7.06*	N/A#	7.02*	5.87*
Tillage	1	30.03***	0.87	N/A	4.55*	1.69
Irrigation	3	0.58	7.31***	N/A	17.80***	22.34***
Cultivar*Tillage	1	0.24	0.16	N/A	0.04	0.17
Cultivar*Irrigation	3	0.31	0.06	N/A	0.60	0.47
Tillage*Irrigation	3	0.72	1.50	N/A	1.35	0.68
Cultivar*Tillage*Irrigation	3	0.12	0.12	N/A	0.45	0.06
Moultrie						
Tillage	1	2.28	0.20	1.72	0.37	4.18
Vienna						
Tillage	1	2.58	12.33**	23.06**	85.48***	44.39***

†Measurements taken at the eight leaf stage (8-lf), early bloom (EB) two weeks after early bloom (EB+2wk), four weeks after early bloom (EB+4wk), and six weeks after early bloom (EB+6).

‡Cultivars included FiberMax 1944 GLB2 (FM 1944) and PhytoGen 499 WRF (PHY 499).

§Tillage treatments included conventional tillage and conservation tillage with a rolled rye cover.

¶Irrigation included 100%, 75%, and 50% of the UGA Checkbook recommended amounts and dryland (unirrigated)

#No plant growth measurements taken at EB+4wk stage in Camilla 2014 trial.

*, **, ***Denotes significant difference at α = 0.05, 0.01, and 0.001, respectively.

Table 4.4. Plant height (cm) as influenced by cultivar, tillage, and irrigation across four locations.

Factor		8-lf†	EB	EB+2 wk	EB+4 wk	EB+6 wk
Camilla 2013						
Cultivar‡	FM 1944	28	52	75	88	96
	PHY 499	30	59	85	102	110
	pLSD ≤ 0.05	1	2	4	5	5
Tillage§	Conv.	30	59	83	102	114
	Rye	29	52	78	88	92
	pLSD ≤ 0.05	1	2	4	5	5
Irrigation¶	100% CHBK	29	59	86	99	105
	75% CHBK	29	58	86	102	114
	50% CHBK	28	54	80	96	104
	Dryland	30	52	68	84	88
	pLSD ≤ 0.05	2	3	5	7	8
Camilla 2014						
Cultivar	FM 1944	31	74	N/A#	83	90
	PHY 499	32	80	N/A	92	100
	pLSD ≤ 0.05	NS	4	N/A	6	8
Tillage	Conv.	34	78	N/A	84	92
	Rye	29	76	N/A	91	97
	pLSD ≤ 0.05	2	NS	N/A	6	NS
Irrigation	100% CHBK	31	80	N/A	96	109
	75% CHBK	31	82	N/A	100	111
	50% CHBK	33	76	N/A	84	87
	Dryland	32	70	N/A	70	72
	pLSD ≤ 0.05	NS	6	N/A	9	11
Moultrie						
Tillage	Conv.	28	72	85	87	84
	Rye	34	74	92	92	92
	pLSD ≤ 0.05	NS	NS	NS	NS	NS
Vienna						
Tillage	Conv.	24	89	106	112	113
	Rye	25	81	92	90	96
	pLSD ≤ 0.05	NS	5	7	6	6

†Measurements taken at the eight leaf stage (8-lf), early bloom (EB) two weeks after early bloom (EB+2wk), four weeks after early bloom (EB+4wk), and six weeks after early bloom (EB+6).

‡Cultivars included FiberMax 1944 GLB2 (FM 1944) and PhytoGen 499 WRF (PHY 499).

§Tillage treatments included conventional tillage and conservation tillage with a rolled rye cover.

¶Irrigation included 100%, 75%, and 50% of the UGA Checkbook recommended amounts and dryland (unirrigated).

#No plant growth measurements taken at EB+4wk stage in Camilla 2014 trial.

Table 4.5. Anova results (F-values) of nodes above white flower measurements from Camilla 2013 and 2014, Moultrie 2014, and Vienna 2014 locations. Factors include cultivar, tillage, irrigation, two way interactions of cultivar by tillage, cultivar by irrigation, and tillage by irrigation, and the three way interaction of cultivar by tillage by irrigation.

Factors	df	EB	EB+2 wk	EB+4 wk	EB+6 wk
Camilla 2013					
Cultivar‡	1	8.41**	1.63	0.60	1.26
Tillage§	1	121.40***	17.15***	8.31**	112.42***
Irrigation¶	3	7.24***	23.22***	3.86*	3.90*
Cultivar*Tillage	1	2.85	0.66	0.28	0.05
Cultivar*Irrigation	3	0.63	0.39	0.33	0.24
Tillage*Irrigation	3	1.77	2.70	1.58	2.65
Cultivar*Tillage*Irrigation	3	1.06	0.84	0.25	0.84
Camilla 2014					
Cultivar	1	1.82	N/A#	18.64***	3.75
Tillage	1	29.65***	N/A	31.70***	15.21***
Irrigation	3	24.15***	N/A	71.70***	71.19***
Cultivar*Tillage	1	0.05	N/A	0.63	1.74
Cultivar*Irrigation	3	0.12	N/A	0.02	0.26
Tillage*Irrigation	3	1.47	N/A	0.67	2.03
Cultivar*Tillage*Irrigation	3	0.05	N/A	0.64	0.52
Moultrie					
Tillage	1	1.04	0.76	0.07	0.60
Vienna					
Tillage	1	1.61	11.51**	18.69**	N/A††

†Measurements taken at the early bloom (EB) two weeks after early bloom (EB+2wk), four weeks after early bloom (EB+4wk), and six weeks after early bloom (EB+6).

‡Cultivars included FiberMax 1944 GLB2 (FM 1944) and PhytoGen 499 WRF (PHY 499).

§Tillage treatments included conventional tillage and conservation tillage with a rolled rye cover.

¶Irrigation included 100%, 75%, and 50% of the UGA Checkbook recommended amounts and dryland (unirrigated).

#No plant growth measurements taken at EB+4wk stage in Camilla 2014 trial.

††No nodes above the uppermost first position white flower at the EB+6wk date at the Vienna 2014 trial.

*, **, ***Denotes significant difference at $\alpha = 0.05$, 0.01, and 0.001, respectively.

Table 4.6. Nodes above white flower (NAWF) as influenced by cultivar, tillage, and irrigation across four locations.

Factor		EB†	EB+2 wk	EB+4 wk	EB+6 wk
Camilla 2013					
Cultivar‡	FM 1944	7.2	7.5	5.3	2.7
	PHY 499	6.8	7.7	5.4	3.0
	pLSD ≤ 0.05	0.3	NS	NS	NS
Tillage§	Conv.	7.9	7.2	5.6	3.9
	Rye	6.2	7.9	5.0	1.8
	pLSD ≤ 0.05	0.3	0.3	0.4	0.4
Irrigation¶	100% CHBK	7.5	8.1	5.1	2.4
	75% CHBK	6.9	8.0	5.9	3.0
	50% CHBK	7.2	7.7	5.3	2.7
	Dryland	6.5	6.5	5.0	3.3
	pLSD ≤ 0.05	0.4	0.4	0.6	0.6
Camilla 2014					
Cultivar	FM 1944	6.9	N/A#	2.4	1.9
	PHY 499	7.1	N/A	3.1	2.3
	pLSD ≤ 0.05	NS	N/A	0.4	NS
Tillage	Conv.	6.6	N/A	2.3	1.7
	Rye	7.4	N/A	3.2	2.5
	pLSD ≤ 0.05	0.3	N/A	0.4	0.4
Irrigation	100% CHBK	7.5	N/A	3.8	4.1
	75% CHBK	7.6	N/A	3.9	2.9
	50% CHBK	6.8	N/A	2.5	1.3
	Dryland	6.2	N/A	0.8	0.1
	pLSD ≤ 0.05	0.4	N/A	0.5	0.6
Moultrie					
Tillage	Conv.	7.9	5.8	2.5	0.3
	Rye	8.2	6.1	2.4	0.5
	pLSD ≤ 0.05	NS	NS	NS	NS
Vienna					
Tillage	Conv.	6.8	6.0	3.4	0.0
	Rye	6.4	5.1	2.5	0.0
	pLSD ≤ 0.05	NS	0.6	0.4	NS

†Measurements taken at the early bloom (EB) two weeks after early bloom (EB+2wk), four weeks after early bloom (EB+4wk), and six weeks after early bloom (EB+6).

‡Cultivars included FiberMax 1944 GLB2 (FM 1944) and PhytoGen 499 WRF (PHY 499).

§Tillage treatments included conventional tillage and conservation tillage with a rolled rye cover.

¶Irrigation included 100%, 75%, and 50% of the UGA Checkbook recommended amounts and dryland (unirrigated).

#No plant growth measurements taken at EB+4wk stage in Camilla 2014 trial.

Table 4.7. Anova results (F-values) of plant mapping measurements from Camilla 2013 and 2014, Moultrie 2014, and Vienna 2014 locations. Factors include cultivar, tillage, irrigation, two way interactions of cultivar by tillage, cultivar by irrigation, and tillage by irrigation, and the three way interaction of cultivar by tillage by irrigation.

Factors	df	Heights	Total Nodes	Total Bolls
Camilla 2013				
Cultivar‡	1	8.83**	5.52*	3.16
Tillage§	1	57.96***	125.99***	95.10***
Irrigation¶	3	9.24***	2.57	1.12
Cultivar*Tillage	1	1.95	0.27	0.14
Cultivar*Irrigation	3	0.54	0.03	0.19
Tillage*Irrigation	3	1.19	1.12	0.94
Cultivar*Tillage*Irrigation	3	0.18	0.42	0.11
Camilla 2014				
Cultivar	1	8.48**	0.35	5.07*
Tillage	1	3.17	10.78**	0.37
Irrigation	3	25.60***	73.70***	31.66***
Cultivar*Tillage	1	0.01	1.63	0.97
Cultivar*Irrigation	3	0.27	0.43	0.28
Tillage*Irrigation	3	1.61	3.60	3.31
Cultivar*Tillage*Irrigation	3	0.05	0.25	0.08
Moultrie				
Tillage	1	5.20	0.83	3.40
Vienna				
Tillage	1	96.09***	10.27*	0.27

†Measurements taken include plant height (Heights), total mainstem nodes (Total Nodes), and total bolls located on reproductive branches (Total Bolls) at the end of the season.

‡Cultivars included FiberMax 1944 GLB2 (FM 1944) and PhytoGen 499 WRF (PHY 499).

§Tillage treatments included conventional tillage and conservation tillage with a rolled rye cover.

¶Irrigation included 100%, 75%, and 50% of the UGA Checkbook recommended amounts and dryland (unirrigated).

*, **, ***Denotes significant difference at $\alpha = 0.05$, 0.01, and 0.001, respectively.

Table 4.8. Plant mapping measurements as influenced by cultivar, tillage, and irrigation across four locations.

Factor		Heights†	Total Nodes	Total Bolls
Camilla 2013				
Cultivar‡	FM 1944	37.2	21.5	85.1
	PHY 499	40.5	20.5	78.1
	pLSD $\leq$ 0.05	2.3	0.8	NS
Tillage§	Conv.	43.1	23.3	100.8
	Rye	34.6	18.7	62.4
	pLSD $\leq$ 0.05	2.3	0.8	8.0
Irrigation¶	100% CHBK	39.8	21.0	82.3
	75% CHBK	42.3	21.9	81.5
	50% CHBK	39.2	20.8	86.4
	Dryland	34.2	20.3	76.3
	pLSD $\leq$ 0.05	3.2	NS	NS
Camilla 2014				
Cultivar	FM 1944	35.5	19.3	77.9
	PHY 499	39.3	19.5	88.3
	pLSD $\leq$ 0.05	2.7	NS	9.4
Tillage	Conv.	36.3	18.9	81.7
	Rye	38.6	20.0	84.5
	pLSD $\leq$ 0.05	NS	0.7	NS
Irrigation	100% CHBK	44.1	22.6	110.1
	75% CHBK	41.8	21.1	98.0
	50% CHBK	34.0	18.1	71.9
	Dryland	29.8	15.9	52.4
	pLSD $\leq$ 0.05	3.8	1.0	13.3
Moultrie				
Tillage	Conv.	34.6	19.7	74.0
	Rye	38.5	20.5	92.5
	pLSD $\leq$ 0.05	NS	NS	NS
Vienna				
Tillage	Conv.	44.8	20.4	79.2
	Rye	37.9	19.8	83.2
	pLSD $\leq$ 0.05	1.6	0.5	NS

†Measurements taken include plant height (Heights), total mainstem nodes (Total Nodes), and total bolls located on reproductive branches (Total Bolls) at the end of the season.
‡Cultivars included FiberMax 1944 GLB2 (FM 1944) and PhytoGen 499 WRF (PHY 499).
§Tillage treatments included conventional tillage and conservation tillage with a rolled rye cover.
¶Irrigation included 100%, 75%, and 50% of the UGA Checkbook recommended amounts and dryland (unirrigated).

Table 4.9. Anova results (F-values) of yield measurements from Camilla 2013 and 2014, Moultrie 2014, and Vienna 2014 locations. Factors include cultivar, tillage, irrigation, two way interactions of cultivar by tillage, cultivar by irrigation, and tillage by irrigation, and the three way interaction of cultivar by tillage by irrigation.

Factors	df	Seedcotton (kg ha^{-1})	Lint Percentage (%)	Lint Yield (kg ha^{-1})
Camilla 2013				
Cultivar‡	1	2.65	174.25***	11.12**
Tillage§	1	27.19***	35.30***	17.51***
Irrigation¶	3	0.90	5.02**	0.80
Cultivar*Tillage	1	0.07	4.16	0.00
Cultivar*Irrigation	3	0.38	0.59	0.26
Tillage*Irrigation	3	1.93	0.24	1.46
Cultivar*Tillage*Irrigation	3	0.10	0.09	0.14
Camilla 2014				
Cultivar	1	17.20***	612.63***	39.64***
Tillage	1	1.97	7.44*	2.56
Irrigation	3	35.62***	19.68***	35.07***
Cultivar*Tillage	1	0.90	0.00	0.69
Cultivar*Irrigation	3	0.74	0.96	1.14
Tillage*Irrigation	3	2.39	1.93	2.46
Cultivar*Tillage*Irrigation	3	0.59	1.21	0.55
Moultrie				
Tillage	1	1.39	N/A#	2.77
Vienna				
Tillage	1	1.92	N/A	0.66

†Measurements taken include plant height (Heights), total mainstem nodes (Total Nodes), and total bolls located on reproductive branches (Total Bolls) at the end of the season.

‡Cultivars included FiberMax 1944 GLB2 (FM 1944) and PhytoGen 499 WRF (PHY 499).

§Tillage treatments included conventional tillage and conservation tillage with a rolled rye cover.

¶Irrigation included 100%, 75%, and 50% of the UGA Checkbook recommended amounts and dryland (unirrigated).

#Lint percentage measurements not replicated at Moultrie 2014 and Vienna 2014 trails.

*, **, ***Denotes significant difference at $\alpha = 0.05$, 0.01, and 0.001, respectively.

Table 4.10. Yield parameters as influenced by cultivar, tillage, and irrigation across four locations.

Factor		Seedcotton (kg ha^{-1})	Lint Percentage (%)	Lint Yield (kg ha^{-1})
Camilla 2013				
Cultivar†	FM 1944	3535.62	38.0	1339.70
	PHY 499	3789.16	41.2	1547.42
	pLSD $\leq$ 0.05	NS	0.5	127.40
Tillage‡	Conv.	4068.31	38.8	1573.86
	Rye	3256.46	40.3	1313.26
	pLSD $\leq$ 0.05	318.00	0.5	127.40
Irrigation§	100% CHBK	3481.74	40.1	1392.53
	75% CHBK	3840.39	39.5	1517.26
	50% CHBK	3686.68	39.8	1451.72
	Dryland	3640.72	38.8	1412.73
	pLSD $\leq$ 0.05	NS	0.7	NS
Camilla 2014				
Cultivar	FM 1944	2620.54	35.7	945.31
	PHY 499	3183.37	40.5	1295.90
	pLSD $\leq$ 0.05	277.14	0.4	113.73
Tillage	Conv.	2997.27	38.4	1165.18
	Rye	2806.64	37.9	1076.03
	pLSD $\leq$ 0.05	NS	0.4	NS
Irrigation	100% CHBK	3198.44	38.9	1256.93
	75% CHBK	3612.85	38.6	1405.77
	50% CHBK	3056.79	38.0	1172.12
	Dryland	1739.74	37.0	647.59
	pLSD $\leq$ 0.05	391.94	0.6	160.84
Moultrie				
Tillage	Conv.	3132.4	N/A#	1187.54
	Rye	2932.4	N/A	1081.18
	pLSD $\leq$ 0.05	NS	N/A	NS
Vienna				
Tillage	Conv.	1323.72	N/A	535.265
	Rye	1291.82	N/A	527.712
	pLSD $\leq$ 0.05	NS	N/A	NS

†Cultivars included FiberMax 1944 GLB2 (FM 1944) and PhytoGen 499 WRF (PHY 499).

‡Tillage treatments included conventional tillage and conservation tillage with a rolled rye cover.

§Irrigation included 100%, 75%, and 50% of the UGA Checkbook recommended amounts and dryland (unirrigated).

#Lint percentage measurements not replicated at Moultrie 2014 and Vienna 2014 trails.

Table 4.11. Anova results (F-values) of fiber quality measurements from Camilla 2013 and 2014, Moultrie 2014, and Vienna 2014 locations. Factors include cultivar, tillage, irrigation, two way interactions of cultivar by tillage, cultivar by irrigation, and tillage by irrigation, and the three way interaction of cultivar by tillage by irrigation.

Factors	df	Staple	Micronaire	Strength	Uniformity
Camilla 2013					
Cultivar‡	1	44.26***	27.66***	0.12	17.92***
Tillage§	1	19.00***	9.73**	8.83**	3.11
Irrigation¶	3	6.09**	2.44	4.80**	1.02
Cultivar*Tillage	1	1.32	10.88**	2.36	0.45
Cultivar*Irrigation	3	1.74	0.47	3.75*	0.48
Tillage*Irrigation	3	0.89	0.94	4.04*	1.91
Cultivar*Tillage*Irrigation	3	0.33	0.60	0.77	3.10*
Camilla 2014					
Cultivar	1	107.23***	49.34***	0.79	23.68***
Tillage	1	5.87*	20.45***	0.02	0.01
Irrigation	3	6.65**	15.29***	0.28	2.69
Cultivar*Tillage	1	0.05	2.08	0.22	0.18
Cultivar*Irrigation	3	1.34	1.94	0.93	1.38
Tillage*Irrigation	3	1.73	3.52*	0.60	1.43
Cultivar*Tillage*Irrigation	3	1.34	0.26	1.32	1.99

†Measurements taken at the eight leaf stage (8-lf), early bloom (EB) two weeks after early bloom (EB+2wk), four weeks after early bloom (EB+4wk), and six weeks after early bloom (EB+6).

‡Cultivars included FiberMax 1944 GLB2 (FM 1944) and PhytoGen 499 WRF (PHY 499).

§Tillage treatments included conventional tillage and conservation tillage with a rolled rye cover.

¶Irrigation included 100%, 75%, and 50% of the UGA Checkbook recommended amounts and dryland (unirrigated)

*, **, ***Denotes significant difference at α = 0.05, 0.01, and 0.001, respectively.

Table 4.12. Fiber quality parameters as influenced by cultivar, tillage, and irrigation across four locations.

Factor		Staple (32^{nds} of an inch)	Micronaire	Strength (g/tex)	Uniformity (%)
Camilla 2013					
Cultivar‡	FM 1944	37.6	4.8	32.0	82.5
	PHY 499	36.4	5.1	32.0	83.4
	pLSD $\leq$ 0.05	0.4	0.1	NS	0.4
Tillage§	Conv.	37.4	4.9	32.4	83.1
	Rye	36.6	5.0	31.6	82.8
	pLSD $\leq$ 0.05	0.4	0.1	0.6	NS
Irrigation¶	100% CHBK	36.8	5.0	31.5	83.0
	75% CHBK	36.6	4.9	31.6	83.2
	50% CHBK	37.2	4.8	32.1	82.8
	Dryland	37.6	5.0	32.8	82.7
	pLSD $\leq$ 0.05	0.5	NS	0.8	NS
Camilla 2014					
Cultivar	FM 1944	38.0	4.2	32.5	82.9
	PHY 499	36.1	4.5	32.9	83.9
	pLSD $\leq$ 0.05	0.4	0.1	NS	0.4
Tillage	Conv.	37.3	4.4	32.7	83.4
	Rye	36.8	4.2	32.6	83.4
	pLSD $\leq$ 0.05	0.4	0.1	NS	NS
Irrigation	100% CHBK	37.3	4.6	32.4	83.6
	75% CHBK	37.6	4.3	32.9	83.7
	50% CHBK	36.8	4.2	32.8	83.3
	Dryland	36.5	4.2	32.6	83.0
	pLSD $\leq$ 0.05	0.5	0.1	NS	NS

†Measurements taken at the eight leaf stage (8-lf), early bloom (EB) two weeks after early bloom (EB+2wk), four weeks after early bloom (EB+4wk), and six weeks after early bloom (EB+6).

‡Cultivars included FiberMax 1944 GLB2 (FM 1944) and PhytoGen 499 WRF (PHY 499).

§Tillage treatments included conventional tillage and conservation tillage with a rolled rye cover.

¶Irrigation included 100%, 75%, and 50% of the UGA Checkbook recommended amounts and dryland (unirrigated).

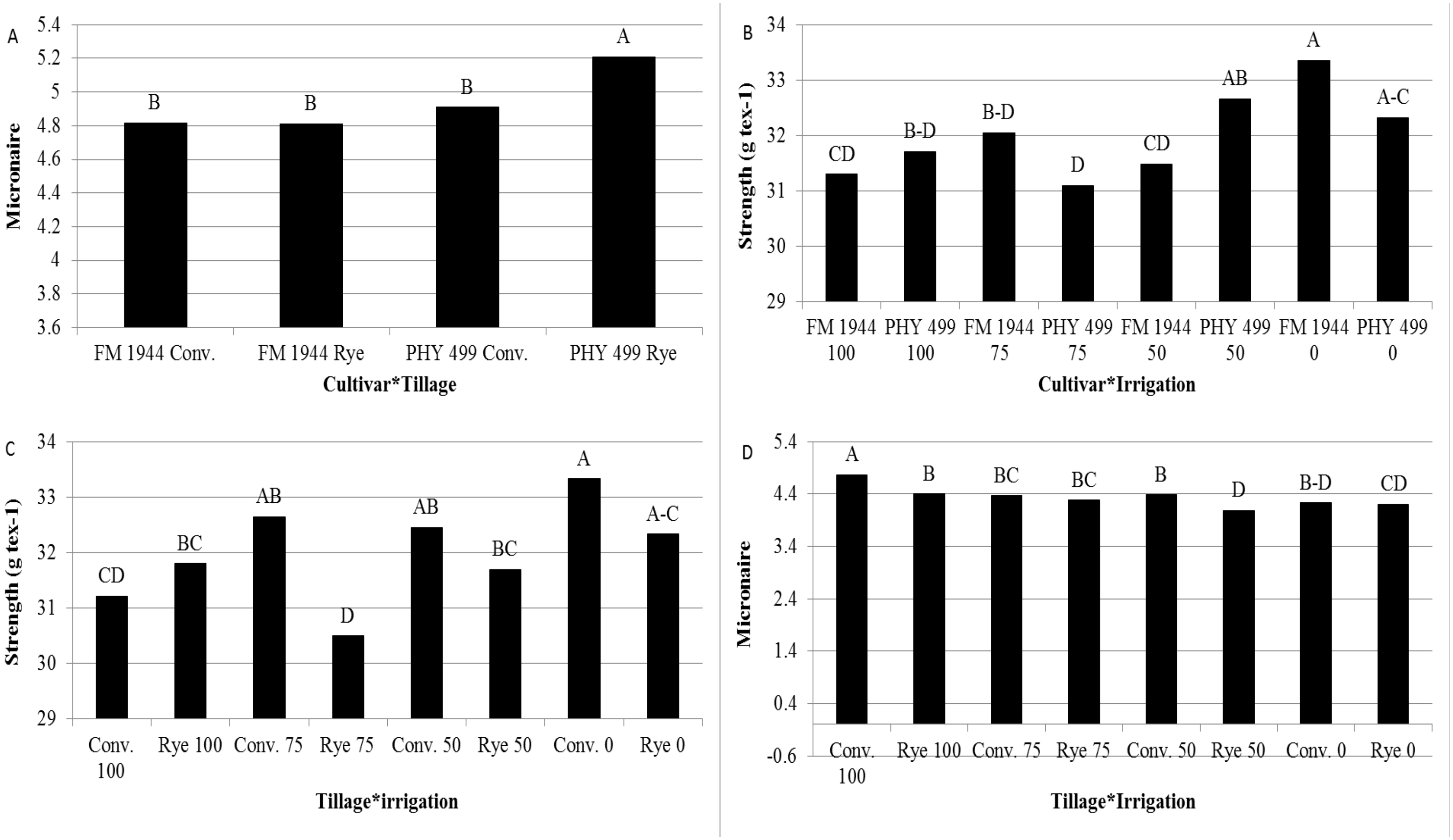

Figure 4.1. The two-way interactions of cultivar by tillage on micronaire at Camilla 2013 (A), cultivar by irrigation on fiber strength (strength) at Camilla 2013 (B), tillage by irrigation on fiber strength (strength) at Camilla 2013 (C), and tillage by irrigation on micronaire at Camilla 2014 (D). Cultivar treatments include FiberMax 1944 GLB2 (FM 1944) and PhytoGen 499 WRF (PHY 499); tillage treatments include conventional (Conv.) and conservation (Rye) tillage; irrigation treatments include 100, 75, and 50% of UGA Checkbook recommendations and dryland (0).

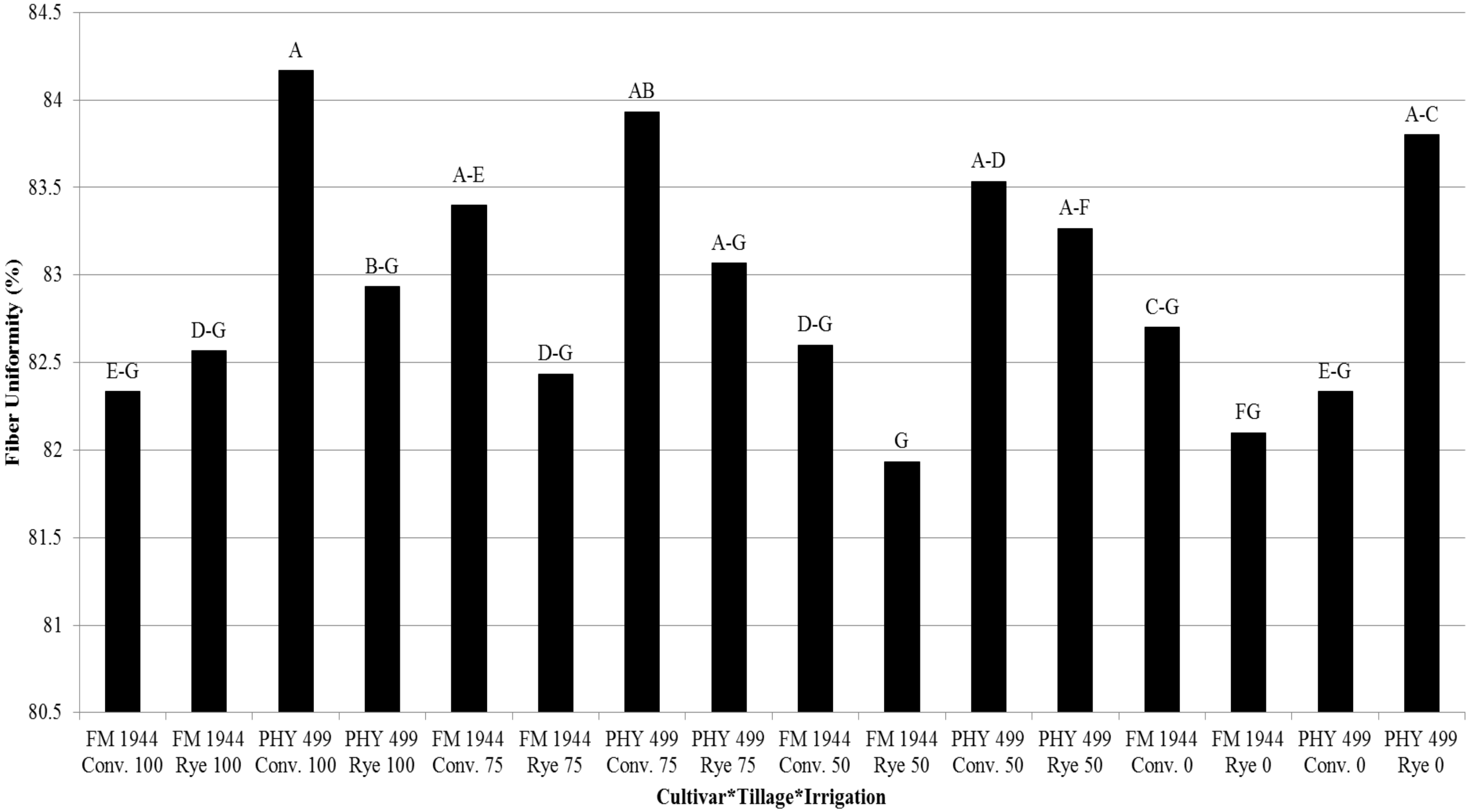

Figure 4.2. The effect of the three-way interaction of cultivar by tillage by irrigation on fiber uniformity at Camilla 2013. Cultivar treatments include FiberMax 1944 GLB2 (FM 1944) and PhytoGen 499 WRF (PHY 499); tillage treatments include conventional (Conv.) and conservation (Rye) tillage; irrigation treatments include 100, 75, and 50% of UGA Checkbook recommendations and dryland (0).

CHAPTER 5

PHYSIOLOGICAL RESPONSES OF COTTON (*GOSSYPIUM HIRSUTUM*) TO WATER DEFICIT AND THE EFFECT OF COVER CROPS ON COTTON WATER STATUS[1]

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[1] S.A. Byrd, J.L. Snider, G.D. Collins, A.S. Culpepper, P.M. Roberts, W.M. Porter, and J.R. Whitaker. To be submitted to *Agronomy Journal*.

Abstract

As the most limiting factor to crop production, the effect of water deficit to physiological crop performance has been and continues to be extensively studied. Cover crops have been observed to have several beneficial effects to crop production, including the increase of soil moisture and improved water retention, but there is limited data about the physiological impact of cover crops on cotton specifically. This study aimed to evaluate the physiological impact of cotton grown under conservation tillage utilizing a rolled rye cover crop compared to conventional tillage under fully irrigated and nonirrigated (dryland) conditions in Georgia. Measurements of fluorescence, gas exchange, photosynthesis, and leaf water potential were conducted on five occasions across 2013 and 2014. When water deficit conditions were present in the dryland treatments, declines in stomatal conductance coincided with reductions in leaf water potential compared to the fully irrigated treatment. At the most severe water deficit period, these declines resulted in reduced photosynthetic rates. Except for one instance of increased leaf water potential, conservation tillage had no effect on the physiological parameters measured compared to conventional tillage. The results of this study mirror previous findings of reduced leaf water potential, stomatal conductance, and photosynthetic rates when plants are under water deficit. In regards to the impact of tillage, the results indicate that conservation tillage utilizing a rolled cover crop has little, if any, benefit to crop performance from a physiological perspective.

Introduction

Plants experience water deficit stress when water lost through transpiration exceeds water uptake (Bray, 1997; Lawlor and Tezara, 2009), which is detrimental to crop growth and physiological processes (Hsiao and Acevedo, 1974; Chaves et al., 2003; Saibo, 2008). During periods of water deficit, inhibition of growth occurs through the reduction in cell expansion and division (Hsiao, 1973). In cotton this reduction in growth due to water deficits, occurring throughout the season or during the flowering period, negatively impacts the development of potential reproductive sites through the reduction of plant height (Grimes et al., 1970; Grimes and Yamada, 1982; Turner et al., 1986; Pettigrew, 2004; Gwathmey et al., 2011), mainstem nodes (Pace et al., 1999; Pettigrew 2004; Gwathmey et al., 2011), and blooms or boll numbers (Grimes et al., 1970; Guinn and Mauney, 1984; Turner et al., 1986; Morrow and Krieg, 1990; Pace et al., 1999; Pettigrew, 2004; Ritchie et al., 2009). Detrimental impacts on leaf area (Grimes et al., 1970; Turner et al., 1986; Gerik et al., 1996; Pace et al., 1999; Carmo-Silva et al., 2012), as well as increases in fruit shedding (Turner et al., 1986) result from water deficit in cotton.

When severe enough, water deficit in cotton inhibits net photosynthesis (Ackerson and Hebert, 1981; Turner et al., 1986; Ratnayaka et al., 2003; Carmo-Silva et al., 2012), reduces the maximum quantum yield of photosystem II (F_v/F_m) (Ratnayaka et al., 2003; Carmo-Silva et al., 2012), and decreases the actual quantum yield of photosystem II (Φ_{PSII}) (Carmo-Silva et al., 2012). However, multiple studies have reported either no effect or an increase in F_v/F_m and Φ_{PSII} resulting from water deficit in cotton (Kitao and Lei, 2007; Massacci et al., 2008; Snider et al., 2014; Chastain et al., 2014). Similar results have been observed for photosynthetic electron transport rate (ETR), as Massacci et al. (2008) and Carmo-Silva et al. (2012) reported a decrease in ETR, while multiple other studies report either no effect or an increase in ETR (Kitao and Lei, 2007; Zhang et al., 2011; Snider et al., 2014; Chastain et al., 2014) under water deficit conditions. Dark respiration (R_D) has been observed to either increase or decrease during periods of water stress (Massacci et al., 2008; Zhang et al., 2011; Chastain et al., 2014), while photorespiration has been reported to either increase (Cornic and Fresneau, 2002; Massacci et al., 2008) or decrease (Perry et al., 1983; Zhang et al., 2011) under water deficit conditions. Stomatal conductance (g_s) is extremely sensitive to water deficit, and this parameter is most directly related to declines in photosynthetic rates in response to water deficit (Medrano et al., 2002; Flexas et al., 2002; Snider et al., 2014). Typically drought-induced declines in g_s are closely associated with declines in leaf water potential (Ψ_L) in cotton (Radin, 1984; Turner et al., 1986; Snider et al., 2014). Whether measured during predawn or afternoon hours, Ψ_L is a direct measure of plant water status that is widely used as an indicator of drought stress and is highly predictive of physiological responses to water deficit (McMichael et al., 1973; Ackerson et al., 1977a; Ackerson et al., 1977b; Ackerson and Hebert, 1981; Radin, 1984; Turner et al., 1986; Snider et al., 2014; Chastain et al., 2014).

Along with these crucial physiological processes, water stress increases canopy temperature (Carmo-Silva et al., 2012) which is typically linked with stomatal closure. When comparing dryland (unirrigated or rain-fed) cotton to well-watered or fully irrigated cotton, an increase in leaf temperatures and the difference between leaf and air temperatures has been reported (Chastain et al., 2014).

Recently, primarily due to suppression of weeds (Creamer et al., 1996; Culpepper et al., 2010; Sosnoskie and Culpepper, 2012;) and pests such as root-knot nematodes (Bauer et al., 2010) and thrips (Manley et al., 2003; Olson et al., 2006), producers have increased their adoption of the use of conservation tillage practices, which includes employing a cover crop terminated a few weeks prior to planting the primary crop. This can be accomplished through either rolling the crop to provide residue for soil coverage, or for other winter cover crops, such as wheat, the cover crop is harvested prior to the planting of the primary crop and the remaining stubble provides a degree of soil coverage and residue for the following growing season. Multiple studies have determined that soil moisture or water content is increased under conservation tillage and with the utilization of a cover crop (Blevins et al., 1971; Gantzer and Blake, 1978; Dao, 1993; Daniel et al., 1999b).

Research on the effect of cover crops on physiological processes of the subsequent crop is limited (Al-Darby et al., 1987; Singer et al., 2007), and has primarily been conducted on corn. There does not appear to be any work on the effect of a cover crop on the physiological response of cotton grown under water stressed conditions in the Southeastern United States. With regard to yield, previous studies have shown increases in cotton yield under conservation tillage compared to conventional tillage, although seasonal rainfall or irrigation patterns greatly influence this result (Bordovsky et al., 1994; Bauer and Busscher, 1996; Raper et al., 2000; Wiatrak et al., 2006; Bauer et al., 2010). Yield increases with conservation tillage have been noted in dryland conditions, indicating that cover crops enhance soil moisture retention, leading to increased yields compared to conventional tillage in dryland conditions (Bordovsky et al., 1994; Bauer et al., 2010). Rye (*Secale cereal*) has become a popular choice of cotton growers in the Southeastern U.S. due to high biomass production which allows for more successful suppression of problematic weeds such as palmer amaranth (*Amaranthus palmeri*) (Culpepper et al., 2010; Sosnoskie and Culpepper, 2012).

Periods of water deficit are inevitable during the cotton growing season in the southeastern United States and the increasing focus on, and regulation of agricultural water use may limit the use of irrigation. Cover crops have the potential to alleviate water stress as a result of increased water availability to the plant. The objective of this study was evaluate the physiological responses of cotton to a high residue rye cover crop, compared to conventional tillage, in both irrigated and non-irrigated conditions. A number of plant water status and physiological measures were performed to determine the response of cotton grown under the different tillage and irrigation systems. We hypothesized that the increased levels of soil moisture documented in previous studies with the utilization of cover crops may allow for higher leaf water potential, resulting in higher stomatal conductance and photosynthetic rates relative to conventionally tilled plots when water deficit conditions are present.

Materials and Methods

Research was conducted during the 2013 and 2014 cotton season at the University of Georgia's C.M. Stripling Irrigation Research Park in Camilla, GA (31° 16' 46" N, 84° 17' 48" W). The soil type at this location is a Lucy loamy sand (USDA NRCS 2014a) classified as a loamy, kaolinitic, thermic Arenic Kandiudult and being very deep, well drained, and moderately permeable (USDA NRCS 2014b). Two tillage treatments were included in the study, conventional tillage and a reduced tillage utilizing a rye cover. Wrens Abruzzi rye was drilled in 19 cm rows at a 101 kg/ha seeding rate with a Great Plains 3P606NT grain drill (Great Plains Ag, Salina, KS) each November prior to the following cotton season. The grain drill had been modified to planting rye seed in rows 15 cm apart except where each cotton row would be planted where a 30.5 cm wide spacing was placed. The goal of the rye plan spacing was to ensure the rye cover would not impede the emergence of cotton. The rye was rolled and sprayed with a burndown herbicide consisting of 1.61 L/ha of glyphosate and 0.15 L/ha of flumioxazin approximately two weeks prior to cotton planting. Plots in conventional tillage contained no rye cover and were planted with the same strip tillage implement utilized for planting the conservation tillage plots. A Monosem STD 540 (Monosem Inc. Edwardsville, KS) two row vacuum planter mounted behind a KMC strip tillage implement (Kelly Manufacturing Co. Tifton, GA) to facilitate planting operations in plots containing the rolled rye cover was utilized for cotton planting in both the conservation and conventional tillage plots.

Two irrigation treatments were utilized in the study, consisting of the UGA checkbook recommended irrigation levels (100% checkbook) and a non-irrigated treatment (dryland). The UGA checkbook contains recommended rates of water on a weekly basis throughout the growing season, with the weekly totals varying depending on growth stage (Collins et al., 2015). It is essentially a water balance method, with irrigation used to supplement rainfall to achieve recommended water supply. Irrigation was applied twice per week to adjust for rainfall, though instances of heavy rainfall after the first application resulted in multiple weeks were water supplied exceeded recommendations. Irrigation was applied through an overhead Valley linear irrigation system (Valmont Industries Inc., Omaha, NE). Plots consisted of two 91.5 cm rows 13.7 m in length. In plots containing the rye cover, cotton was planted into the rye gap with a planter that included a strip tillage implement so that cotton emergence was not impeded by the rolled rye cover. The cultivar FiberMax 1944 GLB2 (Bayer CropScience, Research Triangle Park, NC) was planted in both years of the study. Cotton was planted on 29 April in 2013 and on 9 May in 2014. Rainfall totals were 74.5 and 40.9 cm for the 2013 and 2014 growing seasons, respectively.

Physiological measurements were conducted when conditions were conducive to sampling (clear sky conditions). In 2013, measurements were conducted during the final week of squaring (17, June), the first week of bloom (26, June), and during the third week of bloom (18, July). In 2014 measurements were taken during the first week of bloom (8, July) and the third week of bloom (30, July). At each measurement period, two sets of measurements were performed, one during pre-dawn hours (between 0500 and 0645 hours) and one during midday (conducted between 1200 and 1400 hours). Measurements taken during pre-dawn hours included leaf water potential (Ψ_{PD}), the maximum quantum yield of photosystem II (F_v/F_m), abaxial leaf temperature (T_L), and leaf respiration. Midday measurements included leaf water potential (Ψ_{MD}), actual quantum yield (ΦPSII), photosynthetically active radiation (PAR), abaxial leaf temperature, net photosynthetic rate (P_n), stomatal conductance (g_s), transpiration (E), and electron transport rate (ETR).

Water Potential Measurements. Ψ_{PD} and Ψ_{MD} were measured using a PMS Model 615 Pressure Chamber (PMS Instrument Company, Albany, OR). Measurements were done on the uppermost fully expanded subtending leaf, typically on the fourth node below the terminal. This leaf was cut removed from the plant with a box cutting blade by cutting the petiole near the mainstem ensuring that an adequate amount of the petiole was remaining for Ψ_L determination. Approximately two to three seconds elapsed between the removal of the leaf from the plant until it was placed in the chamber and the chamber was pressurized. Data from Ψ_L measurements is expressed in megapascals. Ψ_{PD} values were subtracted from Ψ_{MD} values to determine the difference between predawn and midday leaf water potential ($\Delta\Psi_L$).

Chlorophyll Fluorescence. A Model OS5p flurometer (Opti-Sciences Inc., Hudson, NH) was utilized for measurements of F_v/F_m, ΦPSII, and PAR. F_v/F_m measurements were conducted during predawn hours to ensure the leaves were dark-adapted. F_0 was measured using a low-intensity, red, modulated light source, F_m was determined following exposure to a saturating flash (15,000 μmol m^{-2} s^{-1}) of a 0.8 s duration. F_v/F_m was calculated using the following equation $(F_0 - F_m)/F_m$ as described in Maxwell and Johnson (2000). For midday chlorophyll fluorescence measurements, steady state fluorescence (F_s) was measured under ambient photosynthetically active radiation (PAR). The average PAR at each sample date ranged from 1384 – 2050 μmol m^{-2} s^{-1}, above the reported light saturation point for photosynthesis in cotton (Constable and Rawson, 1980; Krieg and Sung, 1986; Ehleringer and Hammond, 1987; Constable and Oosterhuis, 2010; Wells, 2011; Chastain et al., 2014). Immediately following F_s measurements, F_m' was measured using the multi-flash protocol in which samples are exposed to three consecutive flashes of increasing intensity (2850, 5700, and 8550 μmol m^{-2} s^{-1}) to estimate F_m' when all reaction centers are closed. This is particularly critical when measuring samples that have been adapted to high light conditions (Earl and Ennahli, 2004). Subsequently, actual quantum yield of photosystem II (ΦPSII) was calculated as (F_s – apparent F_m')/F_m'. PAR was measured at each measurement time using chamber sensors on the Li-Cor 6400 Portable Photosynthesis System (Li-Cor Environmental, Lincoln, NE) utilized on the gas exchange measurements described below. ETR was calculated by multiplying ΦPSII x PAR x 0.5 (excitation energy divided between the two photosystems) x 0.84 (leaf absorbance coefficient for C3 plant species) (Flexas et al., 1999).

Gas Exchange. Predawn measurements of respiration, and midday measurements of net photosynthesis (P_N), stomatal conductance (g_s), and transpiration (E), were measured using a Li-Cor 6400 Portable Photosynthesis System (Li-Cor Environmental, Lincoln, NE). All measurements were taken on the uppermost fully expanded leaf, typically the fourth leaf below the terminal. Measurement settings for both predawn and midday measurements were constant; a flow rate of 500 μmol s^{-1}, reference CO_2 concentration was set to 380 ppm, and block temperature was set to match ambient temperature conditions at the time of measurement. Predawn respiration rates were determined following approximately 120 s after the leaf was enclosed in the chamber (once a steady state had been reached). Midday measurements were conducted under the same ambient PAR levels as noted in the chlorophyll fluorescence section, and data were logged when steady-state net photosynthesis had been reached (approximately 60 s after enclosing the leaf in the chamber). Midday dark respiration (R_D) rates were estimated using the Q_{10} relationship described by Valentini et al. (1995). Gross photosynthesis (P_G) was then estimated by the sum of P_n and R_d. Photorespiration (R_I) rates were estimated using the formula described by Valentini et al. (1995).

A randomized complete block design was utilized with three replications each year and a factorial arrangement of treatments within each replication. Tillage (conservation tillage with rye cover and conventional tillage) was the fixed effect, and irrigation (100% checkbook and nonirrigated or dryland) was the random effect. Statistical analysis was performed using SAS 9.4 software (SAS Institute Inc., Cary, NC) with the macro pdmix 800 (Saxton, 1998). Treatment means were separated by Fisher's Protected LSD at an alpha level of $\leq$ 0.05. When data was pooled, the date of measurement was significant for all physiological parameters measured (data not shown), thus irrigation and tillage main effects were analyzed within each sample date.

Results and Discussion

Meteorological conditions, including PAR, averaged over the span of the midday measurement period from all three dates are presented in Table 5.1. Average PAR values, as described in the materials and methods, ranged from 1384 – 2050 μmol m^{-2} s^{-1}, above the light saturation intensities for net photosynthesis in cotton (Constable and Rawson, 1980; Krieg and Sung, 1986; Ehleringer and Hammond, 1987; Constable and Oosterhuis, 2010; Wells, 2011). Air temperature averages across all measurement dates ranged from 18.7 to 23.8 C for predawn measurements and 29.0 to 32.0 C for midday measurements, with midday temperatures within the optimal range for photosynthesis in cotton (Law and Crafts-Brandner, 1999). Relative humidity was similar across all sample dates within each measurement period. Rainfall during the growing season totaled 76.8 and 36.2 cm for 2013 and 2014, respectively. The irrigated treatment supplemented rainfall with 19.1 cm of irrigation in 2013, and 30.2 cm during 2014. Total rainfall in the seven days preceding the measurement dates were 0.8 cm (all on 10 June) for the 17 June 2013 measurement, 0.9 cm (0.8 on 23, June) for the 26 June 2013 measurement, 2.0 cm (only, 0.03 after 14 July) for the 18 July 2013 measurement, 0 cm for the 8 July 2014 measurement, and 0.15 cm for the 30 July 2014 measurement.

Leaf Water Potential. There was a significant effect of irrigation on three dates (Table 5.2) which resulted in reduced Ψ_{PD} in the dryland treatment (Table 5.3). The dryland treatment contained Ψ_{PD} values 0.04, 0.2, and 0.39 MPa lower than the irrigated treatment on 17 June 2013, 26 June 2013, and 8 July 2014, respectively. The conventionally tilled treatment also reduced Ψ_{PD} 0.1 MPA compared to the conservation tillage treatment on the 17 June 2013 date. On 30 July 2014 the interaction of irrigation and tillage was significant for Ψ_{PD}, with levels significantly lower in the dryland treatment under conventional tillage than in the conservation tillage dryland treatment, but higher in both the irrigated treatments regardless of tillage (Fig. 5.1). The dryland treatment resulted in lower Ψ_{MD} at the 26 June 2013, 8 July and 30 July 2014 measurement dates by 0.72, 0.56, and 1.29 MPa, respectively. The dryland treatment resulted in a greater $\Delta\Psi_L$ at the 26 June 2013 and 30 July 2014 dates, while $\Delta\Psi_L$ was increased due to conservation tillage as compared to conventional tillage at the 18 July 2013 measurement. These instances may illustrate a more rapid decline in plant available water, particularly the multiple occurrences due to the dryland treatment. When significant differences were present, the dryland treatment consistently resulted in lower Ψ_L across both measurement periods and dates, while conventional tillage resulted in lower Ψ_L compared to conservation tillage, though differences between tillage treatments were rare. Both Ψ_{PD} and Ψ_{MD} values reached levels that have been observed in previous studies to result in decreases in physiological parameters such as g_s, E, and P_N (Pettigrew, 2004; Snider et al., 2013b, Chastain et al., 2014; Snider et al., 2014). Reduced growth, leaf wilting, and decreased lint yield (Grimes and Yamada, 1982) as well as increases in leaf and boll abscission (McMichael et al., 1973) have also been reported at Ψ_{PD} and Ψ_{MD} values higher than those reported in the current study.

Chlorophyll Fluorescence. A significant interaction between tillage and irrigation was observed for F_v/F_m on the 26 June 2013 measurement date (Table 5.4) with values in the conventionally tilled dryland treatment greater than all other treatments (Table 5.5, Fig. 5.2). There was no significant effect of irrigation or tillage on ΦPSII at any measurement date. The main effect of irrigation was significant for ETR as the dryland treatment resulted in increased ETR on the 26 June measurement date in 2013. While a significant difference was present at one measurement date for F_v/F_m, the values reported do not reflect stress or even biological significance, as it has been observed that F_v/F_m values below 0.8 are indicative of inhibition of photosynthetic processes (Pettigrew, 2004; Burke, 2007; Massacci et al., 2008; Snider et al., 2013). None of the F_v/F_m values in the current study fell below this 0.8 threshold, agreeing with findings of previous studies that observed either no response or an increase in F_v/F_m in cotton under water deficit conditions (Kitao and Lei, 2007; Massacci et al., 2008; Snider et al., 2013, 2015; Chastain et al., 2014). The lack of significant differences in ΦPSII again agrees with previous findings in regard to water deficit (Kitao and Lei, 2007; Massacci et al., 2008; Snider et al., 2014; Chastain et al., 2014). The one instance of increased ETR in the dryland treatment, with no other instances of significant differences, is in agreement with the findings of previous studies reporting no change or an increase in ETR under a water deficit (Kitao and Lei, 2007; Zhang et al., 2011; Snider et al., 2014; Chastain et al., 2014).

Gas Exchange. The main effect of irrigation was significant for g_s, E, and P_N (Table 5.6), with values of all parameters reduced in the dryland treatment. For g_s, the dryland treatment resulted in a 61, 52, and 73% reduction in rates at the 26 June 2013, 8 July, and 30 July 2014 measurements, respectively (Table 5.7). A 30 and 61% reduction in E rates in the dryland treatment were observed on the 26 June 2013 and 30 July 2014 measurements compared to the irrigated treatment. A 51 and 18% reduction in P_N and P_G was observed at 30 July 2014 in the dryland treatment compared to irrigated values. It is likely not coincidental that the three dates at which significant differences in g_s, E, P_N, and P_G were present were also the only dates at which both Ψ_{PD} and Ψ_{MD} values were reduced due to the dryland treatment. Previous studies have reported P_N to decline with declining g_s (Kitao and Lei, 2007), with sharp declines at g_s rates below either 0.5 (Flexas et al., 2002; Snider et al., 2004) or 0.4 mol H_2O $m^{-2}s^{-1}$ (Medrano et al., 2002; Baker et al., 2007). In the one instance in which P_N was reduced due to the dryland treatment, corresponding g_s rates were 0.71 and 0.19 mol H_2O $m^{-2}s^{-1}$ in the irrigated and dryland treatments, respectively, agreeing with the findings of these previous studies. Previous studies have attributed reduced g_s and P_N accompanied by no change, or an increase in ETR, to an increase in electrons utilized by R_I (Kitao and Lei, 2007; Massacci et al., 2008; Chastain et al., 2014). However, in the current study, while reductions in g_s and P_N were present, the one instance of increased ETR in the dryland treatment occurred on a date when no effect on P_N was observed (26 June 2013). On the 30 July 2014 date when both g_s and P_N were reduced as a result of the dryland treatment there was no effect on ETR. Zhang et al. (2011) hypothesized that under water deficit conditions, nitrogen assimilation in leaves could be an alternate sink for electrons which could explain the lack of effect on R_I at any date in the present study. Irrigation had a significant effect on R_D in 2014, as R_D was increased in the irrigated treatment when measured on 8 July, while R_D was higher in the dryland treatment at the 30 July measurement (Table 5.7). These findings correspond with the findings that R_D can increase or decrease under water stress (Loka et al., 2011; Snider and Oosterhuis, 2015). Previous studies have found varying responses of respiration to drought, with respiration increasing or decreasing depending on species (Flexas et al., 2006; Galmés et al., 2007; Atkin et al., 2009), and the timing of the measurement in regards to the duration of the water deficit, a response that has been observed in cotton (Pallas et al., 1967).

Leaf and Air Temperature. For predawn readings, the dryland treatment on 26 June 2013 had a lower difference between leaf and air temperature ($T_L – T_A$) compared to the irrigated treatment, while T_L was significantly higher in the dryland treatment on this date (Table 5.8). While leaf temperatures across both irrigated and dryland treatments were lower than the air temperature on this date, the irrigated treatment resulted in a greater difference, or lower leaf temperature (Table 5.9). For midday measurements on the 26 June 2013 and 30 July 2014 dates the dryland treatment resulted in an increase in both T_L and $T_L – T_A$ (Table 5.8). On 26 June 2013, leaf temperatures were higher than the air temperature in both the irrigated and dryland treatments, while on 30 July 2014, the irrigated treatment resulted in leaf temperatures lower than the air temperature while leaf temperatures were higher than the air temperature in the dryland treatment (Table 5.9).

The variable water supply (irrigated vs. dryland) was the primary driver of physiological differences among treatments throughout the course of this study, with tillage having an effect to a much lesser extent. The findings of this study support previous observations of water deficits leading to reductions in Ψ_L, g_s, P_N, E, and T_L. When water deficit conditions were severe enough to reduce Ψ_L, g_s was decreased resulting in reductions in P_N, E, and T_L. For many of the parameters measured, differences appeared only in 2014 which is likely due to the high precipitation amount that was received during the 2013 season. Not surprisingly, lint yield was not affected by the irrigation treatments in 2013 providing evidence that significant differences due to irrigation treatment in this year did not impact crop performance in terms of yield. The effect of conservation tillage, while minimal, did show some increases in Ψ_{PD} in the two instances in which it was significant. However, the conservation tillage treatment resulted in a lint yield reduction of 261 kg ha^{-1} (Byrd et al., unpublished data). This is likely in response to overly saturated soil conditions, although the definite cause of yield loss due to conservation tillage in 2013 is unclear, as no evidence of detrimental impacts on plant function were observed through the physiological parameters measured in the current study. Large differences in physiological parameters between irrigation treatments were primarily observed at one measurement date, 30 July 2014. As expected based on in-season observations, in 2014 the dryland treatment exhibited a 609 kg ha^{-1} lint yield decrease compared to the irrigated treatment (Byrd et al., unpublished data). This indicates that the utilization of a cover crop did not appear to improve photosynthetic performance over the wide range of water availability observed in the current study. The lack of significant response to conservation tillage for the numerous parameters quantified in this study shows that a cover crop may provide limited benefits, with greater detriments being observed in one year with regard to lint yield. It appears few, if any studies have evaluated the physiological impact of cover crops to cotton grown under varying water supply scenarios in the southeastern United States. Thus, more work would need to be done encompassing differing environmental conditions and irrigation management practices before concrete conclusions on the potential benefits and drawbacks of cover crops to photosynthetic performance could be made.

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Table 5.1. Meteorological data from the predawn and midday measurement periods during 2013 and 2014.

Measurement Date	PAR†	Predawn Air Temp. (C)	Midday Air Temp. (C)	Predawn Rel. Humidity (%)	Midday Rel. Humidity (%)
17 June, 2013	1384	23.8	30.0	93.4	48.8
26 June, 2013	1743	21.0	31.3	95.5	54.8
18 July, 2013	2012	21.9	30.8	93.8	58.4
8 July, 2014	1798	22.6	32.0	93.4	49.9
30 July, 2014	2050	18.7	29.0	84.5	41.8

†Data includes photosynthetically active radiation (PAR), air temperature (Air Temp.), and relative humidity (Rel. Humidity) during both the predawn and midday measurement periods in each year.

Table 5.2. Anova results (F-values) for leaf water potential measurements in 2013 and 2014. Factors include irrigation, tillage, and the interaction of irrigation by tillage.

Factors	df	Ψ_{PD} (MPa)	Ψ_{MD} (MPa)	$\Delta\Psi_L$ (MPa)
17 June, 2013				
Irrigation‡	1	6.18*	0.96	0.71
Tillage§	1	38.45***	1.91	0.88
Irrigation*Tillage	1	1.20	1.34	2.39
26 June, 2013				
Irrigation	1	15.11**	55.59***	40.37***
Tillage	1	1.40	4.22	2.84
Irrigation*Tillage	1	0.26	2.11	1.92
18 July, 2013				
Irrigation	1	0.00	0.12	0.23
Tillage	1	0.61	3.03	10.32*
Irrigation*Tillage	1	0.61	0.12	0.13
8 July, 2014				
Irrigation	1	17.92**	8.56*	1.35
Tillage	1	1.37	1.13	0.43
Irrigation*Tillage	1	0.11	2.02	4.29
30 July, 2014				
Irrigation	1	107.44***	142.16***	23.90**
Tillage	1	9.33*	0.72	0.45
Irrigation*Tillage	1	5.79*	1.00	0.07

†Measurements include predawn leaf water potential (Ψ_{PD}), midday leaf potential (Ψ_{MD}), and the change in leaf water potential between predawn and midday measurements ($\Delta\Psi_L$).

‡Irrigation treatments include 100% of the UGA Checkbook and dryland.

§Tillage treatments include conventional tillage and conservation tillage (rolled rye cover).

*, **, ***Denotes significant difference at $\alpha = 0.05$, 0.01, and 0.001, respectively.

Table 5.3. Means from the main effects of irrigation and tillage on leaf water potential measurements in 2013 and 2014.

		Ψ_{PD} (MPa)†	Ψ_{MD} (MPa)	$\Delta\Psi_{L}$ (MPa)
Date	**Irrigation**			
17 June, 2013	100% CHBK	-0.34	-1.2	-0.86
	Dryland	-0.38	-1.4	-1.00
	$PLSD_{0.05}$	0.04	NS	NS
	Tillage			
	Conv.	-0.41	-1.40	-1.01
	Rye	-0.31	-1.16	-0.85
	$PLSD_{0.05}$	0.04	NS	NS
26 June, 2013	**Irrigation**			
	100% CHBK	-0.37	-1.00	-0.64
	Dryland	-0.56	-1.73	-1.17
	$PLSD_{0.05}$	0.12	0.23	0.20
	Tillage			
	Conv.	-0.49	-1.47	-0.97
	Rye	-0.43	-1.27	-0.83
	$PLSD_{0.05}$	NS	NS	NS
18 July, 2013	**Irrigation**			
	100% CHBK	-0.25	-1.03	-0.75
	Dryland	-0.25	-1.02	-0.73
	$PLSD_{0.05}$	NS	NS	NS
	Tillage			
	Conv.	-0.27	-0.98	-0.69
	Rye	-0.24	-1.07	-0.80
	$PLSD_{0.05}$	NS	NS	0.08
8 July, 2014	**Irrigation**			
	100% CHBK	-0.73	-1.82	-1.29
	Dryland	-1.12	-2.39	-1.46
	$PLSD_{0.05}$	0.22	0.45	NS
	Tillage			
	Conv.	-0.98	-2.21	-1.43
	Rye	-0.87	-2.00	-1.33
	$PLSD_{0.05}$	NS	NS	NS
30 July, 2014	**Irrigation**			
	100% CHBK	-0.30	-1.00	-0.70
	Dryland	-0.92	-2.29	-1.37
	$PLSD_{0.05}$	0.14	0.26	0.32
	Tillage			
	Conv.	-0.70	-1.69	-0.99
	Rye	-0.52	-1.60	-1.08
	$PLSD_{0.05}$	0.14	NS	NS

†Measurements include predawn leaf water potential (Ψ_{PD}), midday leaf potential (Ψ_{MD}), and the change in leaf water potential between predawn and midday measurements ($\Delta\Psi_{L}$).

NS signifies no significant difference.

Table 5.4. Anova results (F-values) for chlorophyll fluorescence measurements in 2013 and 2014. Factors include irrigation, tillage, and the interaction of irrigation by tillage.

			Trait†	
Factors	df	F_v/F_m	ΦPSII	ETR
17 June, 2013				
Irrigation‡	1	0.04	1.53	0.14
Tillage§	1	4.42	0.79	0.00
Irrigation*Tillage	1	0.27	1.36	1.75
26 June, 2013				
Irrigation	1	35.00***	5.33	9.41*
Tillage	1	19.15**	2.79	2.28
Irrigation*Tillage	1	13.00**	0.85	0.80
18 July, 2013				
Irrigation	1	1.26	1.18	0.91
Tillage	1	0.07	0.94	2.17
Irrigation*Tillage	1	0.00	0.82	0.52
8 July, 2014				
Irrigation	1	4.60	1.15	0.17
Tillage	1	4.32	0.93	0.80
Irrigation*Tillage	1	0.05	0.09	0.04
30 July, 2014				
Irrigation	1	2.61	1.52	0.58
Tillage	1	0.22	0.33	0.19
Irrigation*Tillage	1	4.90	0.14	0.20

†Measurements include photosystem II efficiency (F_v/F_m), quantum yield of photosystem II (ΦPSII), and electron transport rate (ETR).

‡Irrigation treatments include 100% of the UGA Checkbook and dryland.

§Tillage treatments include conventional tillage and conservation tillage (rolled rye cover).

*, **, ***Denotes significant difference at $\alpha = 0.05$, 0.01, and 0.001, respectively.

Table 5.5. Means from the main effects of irrigation and tillage on chlorophyll fluorescence measurements in 2013 and 2014.

		F_v/F_m†	ΦPSII	ETR
Date	**Irrigation**			
17 June, 2013	100% CHBK	0.84	0.46	203.40
	Dryland	0.84	0.48	214.66
	$PLSD_{0.05}$	NS	NS	NS
	Tillage			
	Conv.	0.84	0.47	208.18
	Rye	0.83	0.46	209.89
	$PLSD_{0.05}$	NS	NS	NS
26 June, 2013	**Irrigation**			
	100% CHBK	0.82	0.42	308.27
	Dryland	0.83	0.47	346.87
	$PLSD_{0.05}$	0.01	NS	29.74
	Tillage			
	Conv.	0.83	0.46	337.07
	Rye	0.82	0.43	318.07
	$PLSD_{0.05}$	0.01	NS	NS
18 July, 2013	**Irrigation**			
	100% CHBK	0.82	0.44	371.99
	Dryland	0.82	0.48	399.49
	$PLSD_{0.05}$	NS	NS	NS
	Tillage			
	Conv.	0.82	0.45	364.59
	Rye	0.82	0.47	406.90
	$PLSD_{0.05}$	NS	NS	NS
8 July, 2014	**Irrigation**			
	100% CHBK	0.84	0.55	386.62
	Dryland	0.85	0.58	395.78
	$PLSD_{0.05}$	NS	NS	NS
	Tillage			
	Conv.	0.85	0.55	381.38
	Rye	0.84	0.58	401.01
	$PLSD_{0.05}$	NS	NS	NS
30 July, 2014	**Irrigation**			
	100% CHBK	0.81	0.50	425.70
	Dryland	0.82	0.47	400.70
	$PLSD_{0.05}$	NS	NS	NS
	Tillage			
	Conv.	0.81	0.48	405.98
	Rye	0.81	0.49	420.42
	$PLSD_{0.05}$	NS	NS	NS

†Measurements include photosystem II efficiency (F_v/F_m), quantum yield of photosystem II (ΦPSII), and electron transport rate (ETR).

NS signifies no significant difference.

Table 5.6. Anova results (F-values) for single leaf gas exchange measurements in 2013 and 2014. Factors include irrigation, tillage, and the interaction of irrigation by tillage.

Factors	df	Trait†					
		g_s (mol H_2O m^{-2}s^{-1})	E (mmol H_2O m^{-2}s^{-1})	P_N (µmol m^{-2}s^{-1})	P_G (µmol m^{-2}s^{-1})	R_D (µmol m^{-2}s^{-1})	R_I (µmol m^{-2}s^{-1})
17 June, 2013							
Irrigation‡	1	0.69	1.10	0.64	0.21	0.54	0.54
Tillage§	1	2.02	1.76	1.09	1.24	1.12	0.40
Irrigation*Tillage	1	0.07	0.23	0.16	0.25	0.48	2.33
26 June, 2013							
Irrigation	1	8.29*	6.12*	3.80	0.51	2.91	3.70
Tillage	1	0.04	0.25	0.08	0.12	0.06	0.90
Irrigation*Tillage	1	0.23	0.09	0.00	0.06	0.16	0.34
18 July, 2013							
Irrigation	1	0.00	0.00	0.93	0.04	1.01	0.68
Tillage	1	0.90	1.04	3.29	3.14	0.30	3.38
Irrigation*Tillage	1	3.07	3.72	1.17	1.94	0.84	1.09
8 July, 2014							
Irrigation	1	7.47*	4.38	2.04	3.67	7.59*	1.85
Tillage	1	2.41	1.50	1.53	0.67	1.28	0.22
Irrigation*Tillage	1	3.98	2.93	3.36	3.22	0.57	0.48
30 July, 2014							
Irrigation	1	188.14***	75.39***	36.50***	23.03**	7.59*	0.15
Tillage	1	4.26	3.44	2.48	1.29	0.60	0.02
Irrigation*Tillage	1	0.38	0.11	0.23	1.52	1.05	0.49

†Measurements include stomatal conductance (g_s), transpiration (E), net photosynthesis (P_N), gross photosynthesis (P_G), dark respiration (R_D), and gross photosynthesis (P_G).

‡Irrigation treatments include 100% of the UGA Checkbook and dryland.

§Tillage treatments include conventional tillage and conservation tillage (rolled rye cover).

*, **, ***Denotes significant difference at $\alpha = 0.05$, 0.01, and 0.001, respectively.

Table 5.7. Means from the main effects of irrigation and tillage on single leaf gas exchange measurements in 2013 and 2014.

		g_s† (mol H_2O $m^{-2}s^{-1}$)	E (mmol H_2O $m^{-2}s^{-1}$)	P_N (µmol $m^{-2}s^{-1}$)	P_G (µmol $m^{-2}s^{-1}$)	R_D (µmol $m^{-2}s^{-1}$)	R_I (µmol $m^{-2}s^{-1}$)
Date	**Irrigation‡**						
17 June, 2013	100% CHBK	1.52	17.63	32.33	34.59	2.36	5.81
	Dryland	1.39	16.83	30.54	33.33	2.89	7.17
	PLSD$_{0.05}$	NS	NS	NS	NS	NS	NS
	Tillage§						
	Conv.	1.34	16.73	32.59	35.50	3.00	5.91
	Rye	1.57	17.73	30.27	32.41	2.24	7.08
	PLSD$_{0.05}$	NS	NS	NS	NS	NS	NS
26 June, 2013	**Irrigation**						
	100% CHBK	0.57	15.36	28.14	38.54	10.40	12.84
	Dryland	0.22	10.68	20.23	34.96	14.73	17.25
	PLSD$_{0.05}$	0.28	4.47	NS	NS	NS	NS
	Tillage						
	Conv.	0.41	12.55	23.62	35.87	12.25	16.13
	Rye	0.38	13.50	24.75	37.63	12.88	13.96
	PLSD$_{0.05}$	NS	NS	NS	NS	NS	NS
18 July, 2013	**Irrigation**						
	100% CHBK	0.39	12.89	29.73	35.76	6.03	19.30
	Dryland	0.39	12.89	31.40	36.19	4.79	21.45
	PLSD$_{0.05}$	NS	NS	NS	NS	NS	NS
	Tillage						
	Conv.	0.40	13.04	32.13	37.88	5.75	17.98
	Rye	0.38	12.74	29.00	34.07	5.07	22.78
	PLSD$_{0.05}$	NS	NS	NS	NS	NS	NS
8 July, 2014	**Irrigation**						
	100% CHBK	0.25	8.21	16.84	24.05	6.56	25.70
	Dryland	0.12	5.97	13.35	18.73	4.73	28.24
	PLSD$_{0.05}$	0.11	NS	NS	NS	1.57	NS
	Tillage						
	Conv.	0.15	6.43	13.58	20.26	6.02	26.53
	Rye	0.23	7.74	16.60	22.53	5.27	27.41
	PLSD$_{0.05}$	NS	NS	NS	NS	NS	NS
30 July, 2014	**Irrigation**						

100% CHBK	0.71	20.00	40.47	53.25	12.25	16.29
Dryland	0.19	7.72	19.67	43.60	23.40	17.42
$PLSD_{0.05}$	0.09	3.33	8.14	4.76	9.57	NS
Tillage						
Conv.	0.41	12.53	27.36	47.28	19.39	16.63
Rye	0.49	15.14	32.78	49.56	16.26	17.07
$PLSD_{0.05}$	NS	NS	NS	NS	NS	NS

†Measurements include stomatal conductance (g_s), transpiration (E), net photosynthesis (P_N), gross photosynthesis (P_G), dark respiration (R_D), and photorespiration (R_I).

NS signifies no significant difference.

Table 5.8. Anova results (F-values) for leaf and air temperature measurements in 2013 and 2014. Factors include irrigation, tillage, and the interaction of irrigation by tillage.

		Trait†			
		Predawn		Midday	
Factors	df	T_L (C)	$T_L - T_A$ (C)	T_L (C)	$T_L - T_A$ (C)
17 June, 2013					
Irrigation‡	1	0.58	0.58	0.09	0.09
Tillage§	1	3.95	3.95	0.64	0.64
Irrigation*Tillage	1	5.26	5.26	0.37	0.37
26 June, 2013					
Irrigation	1	6.00*	6.00*	39.08***	39.08***
Tillage	1	3.63	3.63	0.33	0.33
Irrigation*Tillage	1	0.21	0.21	1.07	1.07
18 July, 2013					
Irrigation	1	0.08	0.08	2.34	2.34
Tillage	1	0.02	0.02	0.05	0.05
Irrigation*Tillage	1	0.71	0.71	0.17	0.17
8 July, 2014					
Irrigation	1	0.63	0.63	3.04	3.04
Tillage	1	0.53	0.53	0.10	0.10
Irrigation*Tillage	1	0.02	0.02	0.64	0.64
30 July, 2014					
Irrigation	1	0.04	0.04	105.59***	105.59***
Tillage	1	0.12	0.12	3.04	3.04
Irrigation*Tillage	1	0.02	0.02	1.27	1.27

†Measurements include stomatal conductance (g_s), transpiration (E), net photosynthesis (P_N), gross photosynthesis (P_G), dark respiration (R_D), and gross photosynthesis (P_G).

‡Irrigation treatments include 100% of the UGA Checkbook and dryland.

§Tillage treatments include conventional tillage and conservation tillage (rolled rye cover).

*, **, ***Denotes significant difference at $\alpha = 0.05$, 0.01, and 0.001, respectively.

Table 5.9. Means from the main effects of irrigation and tillage on leaf and air temperature measurements in 2013 and 2014.

		Predawn		Midday	
		T_L†	$T_L - T_A$	T_L	$T_L - T_A$
Date	**Irrigation**				
17 June, 2013	100% CHBK	25.46	1.16	30.72	-1.54
	Dryland	25.37	1.07	30.92	-1.34
	$PLSD_{0.05}$	NS	NS	NS	NS
	Tillage				
	Conv.	25.52	1.22	31.10	-1.16
	Rye	25.31	1.01	30.54	-1.72
	$PLSD_{0.05}$	NS	NS	NS	NS
26 June, 2013	**Irrigation**				
	100% CHBK	21.08	-0.65	33.13	1.29
	Dryland	21.30	-0.43	35.85	4.01
	$PLSD_{0.05}$	0.22	0.22	1.03	1.03
	Tillage				
	Conv.	21.28	-0.45	34.62	2.77
	Rye	21.10	-0.63	34.37	2.52
	$PLSD_{0.05}$	NS	NS	NS	NS
18 July, 2013	**Irrigation**				
	100% CHBK	21.49	-0.81	34.04	2.86
	Dryland	21.46	-0.84	32.77	1.58
	$PLSD_{0.05}$	NS	NS	NS	NS
	Tillage				
	Conv.	21.48	-0.82	33.31	2.13
	Rye	21.47	-0.83	33.50	2.32
	$PLSD_{0.05}$	NS	NS	NS	NS
8 July, 2014	**Irrigation**				
	100% CHBK	22.51	-0.60	34.40	1.95
	Dryland	22.38	-0.73	35.66	3.20
	$PLSD_{0.05}$	NS	NS	NS	NS
	Tillage				
	Conv.	22.51	-0.60	35.14	2.69
	Rye	22.39	-0.73	34.92	2.46
	$PLSD_{0.05}$	NS	NS	NS	NS
30 July, 2014	**Irrigation**				
	100% CHBK	18.40	0.02	28.19	-1.22
	Dryland	18.36	-0.03	33.69	4.28
	$PLSD_{0.05}$	NS	NS	1.27	1.27
	Tillage				
	Conv.	18.42	0.03	31.40	2.00
	Rye	18.34	-0.05	30.47	1.06
	$PLSD_{0.05}$	NS	NS	NS	NS

†Measurements include leaf temperature (T_L) and the difference between leaf and air temperature ($T_L - T_A$).

NS signifies no significant difference.

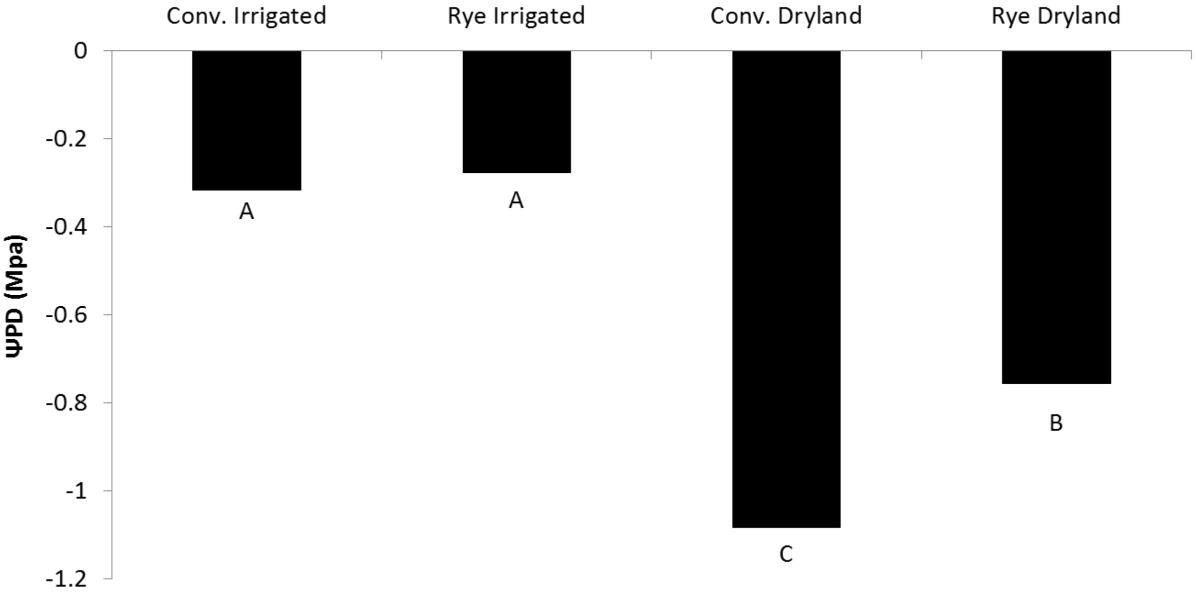

Figure 5.1. Predawn water potential (Ψ_{PD}) on 30 July 2014 in conventional (Conv.) and conservation tillage with a high biomass rye cover crop (Rye) under irrigated and non-irrigated (dryland) conditions.

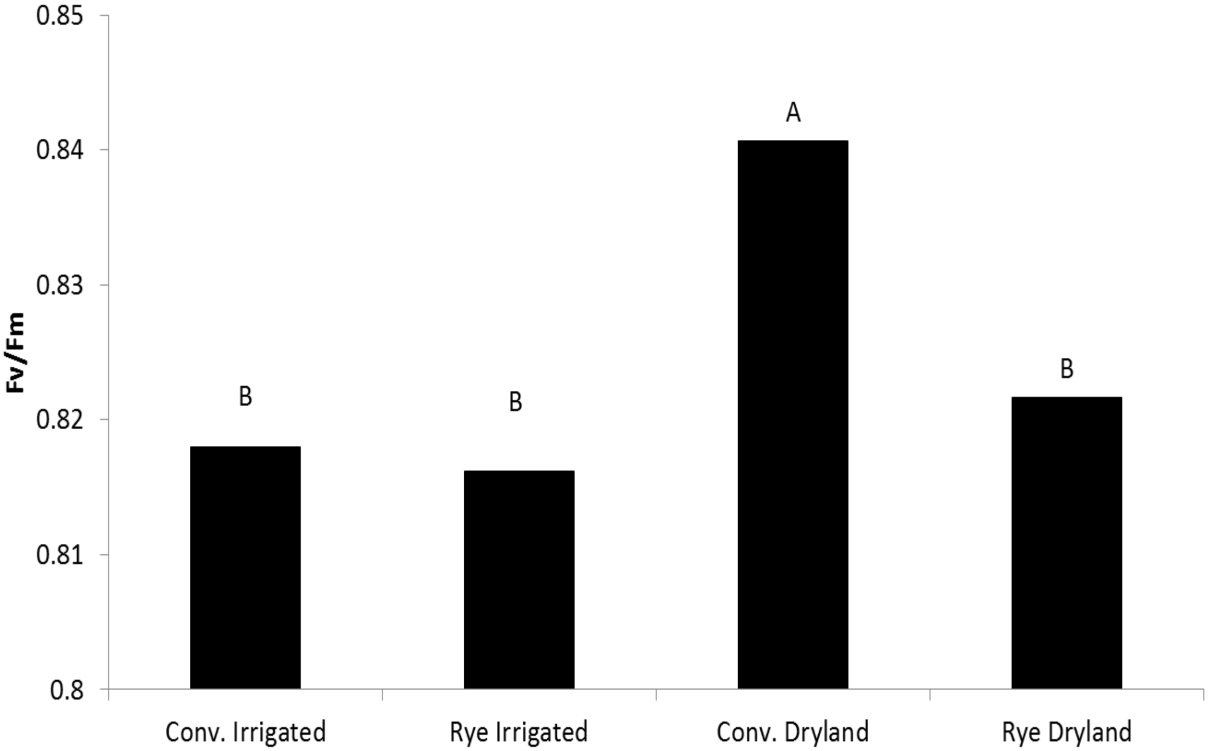

Figure 5.2. Maximum quantum yield of photosystem II (F_v/F_m) on 26 June 2013 in conventional (Conv.) and conservation tillage with a high biomass rye cover crop (Rye) under irrigated and non-irrigated (dryland) conditions.

CHAPTER 6

LEAF PUBESCENCE AND DEFOLIATION STRATEGY INFLUENCE COTTON DEFOLIATION AND FIBER QUALITY[1]

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[1] S.A. Byrd, K.L Edmisten, P.M. Roberts, A.S. Culpepper, J.L. Snider, T.A. Spivey, J.R. Whitaker, W.M. Porter, and G.D. Collins. To be submitted to *Journal of Cotton Science*.

Abstract

Proper defoliation of cotton is critical to maximize both harvest efficiency and fiber quality. Increased levels of leaf or trash grade resulting from inadequate defoliation can lead to decreases in fiber quality and value. Both inherent characteristics of cultivars, such as leaf pubescence levels, as well as defoliation practices influence the efficacy of cotton defoliation. This study aimed to determine the impact of leaf pubescence and defoliation strategies on defoliation success and fiber quality in cotton. Treatments included a factorial of four cultivars and two defoliation treatments. Cultivars included two smooth leaf cultivars and two cultivars with greater leaf pubescence ratings. Defoliation treatments included a standard program and an aggressive program with increased levels of the same defoliant mixture and the addition of a desiccant. There were few instances of a cultivar by defoliation strategy interaction, however, both cultivar and defoliation strategy had a significant effect on defoliation ratings. The aggressive defoliation treatment decreased defoliation and increased desiccation in all three locations but did not influence yield or fiber quality. In two of three locations, cultivars with higher leaf pubescence ratings resulted in increased leaf grades and HVI trash ratings compared with the smooth leaf cultivars. The results of this study conclude that defoliation strategy can impact efficacy of defoliation, while leaf pubescence characteristics influence fiber quality parameters.

Introduction

Application of harvest-aids is often required for producers of indeterminate crops such as cotton. Proper defoliation is critical to maximizing the yield and profitability of the crop. Because cotton is a perennial crop that is grown and managed as an annual for agronomic benefits, eliminating green, live plant material and minimizing the amount of dead plant material contaminating harvested seedcotton is crucial to optimize harvest efficiency and lint quality (Colwick et al., 1984). Proper defoliation has numerous benefits including reducing the amount of leaf and other plant material (referred to as trash) in the harvested seedcotton (Brecke et al., 2001; Valco and Snipes, 2001), reducing damage to fiber in the ginning process by lessening the amount of cleaning required for achieving marketable lint (Valco and Snipes, 2001), reducing losses to boll rot (Brown, 1953), and allowing for earlier harvest to avoid weathering (Cathey et al., 1982; Siebert et al., 2006). While many factors influence the effectiveness of cotton defoliation (Brecke et al., 2001; Siebert et al., 2006), leaf pubescence and defoliation practices are of specific interest with this experiment.

Reduced leaf pubescence has been an important goal of cotton breeding to improve lint quality (Colwick et al., 1984). Leaf pubescence can influence cotton production and ginning practices in regard to reduced defoliation efficacy and reduced lint quality due to increased trash content, as the pubescent plant material can become entangled in the lint. A reduction in motes (Novick et al., 1991), greater lint cleaning efficacy (Colwick et al, 1984; Novick et al., 1991; Bechere et al., 2011), and a reduction in trash or non-lint material in ginned lint has been observed in cultivars with reduced leaf pubescence compared to hairier cultivars (Ramey, 1962; Smith, 1964; Wanjura et al., 1976; Novick et al., 1991).

Defoliants are a common category of harvest-aid products utilized in cotton and achieve leaf removal through the formation of an abscission layer at the base of the petiole (Cathey, 1986) and function optimally when applied to mature, healthy leaves at appropriate rates (Brecke et al., 2001). Plant tissue must be alive for the formation of an abscission layer to occur, thus optimal defoliation could be prevented if cell and tissue death occur too rapidly, inhibiting the formation of the abscission layer (Stahler, 1953; Cathey, 1986; Stichler et al., 1995; Clark and Carpenter, 1997). Leaf removal typically occurs seven to 17 days after the application of defoliants (Colwick et al., 1984; Clark and Carpenter, 1997).

Herbicidal defoliants or desiccants are also used as harvest-aids in cotton. The use of a desiccant as a harvest-aid, can lead to dead leaves remaining on the plant, termed “leaf stick” due to the formation of an abscission layer being inhibited as a result of rapid plant tissue death (Stahler, 1953; McMeans et al., 1966; Bovey and Miller, 1968; Brecke et al., 2001; Shaw, 2002). Leaf stick resulting from desiccation can lead to an increase of trash in ginned lint, as dead leaves are present on the plant at harvest and are removed by the cotton harvester along with the seedcotton (Shaw, 2002). Death of leaves resulting from the utilization of desiccants as a harvest-aid has been observed to occur six to seven days after application (Bovey and Miller, 1968; Clark and Carpenter, 1997).

Increased levels of trash in lint decrease the quality of the lint and could potentially lead to discounted returns to the producer. Both leaf pubescence levels and defoliation strategy have been shown to influence the efficacy of defoliation in cotton, as well as fiber quality characteristics, namely leaf or trash grade. The objective of the current study was to determine the influence of leaf pubescence and defoliation practices on the effectiveness of defoliation and related fiber quality parameters.

Materials and Methods

Experiments were conducted at the University of Georgia’s Gibbs Farm in Tifton, GA (31°26’02.96”N, 83°35’12.48”W) in 2013 and 2014 (Tifton, GA 2013 and Tifton, GA 2014) and at North Carolina State University’s Peanut Belt Research Station in Lewiston, NC (36°07’59.68”N, 77°10’14.06”W) in 2014 (Lewiston, NC 2014). Planting occurred on 25 April, 2013 and 28 April, 2014 in Tifton, at a seeding rate of 11 seeds m^{-1}. Cotton was planted on 12 May, 2014 at the Lewiston location at a rate of 10 seeds m^{-1}. Plot lengths at all locations were 9 m and contained four rows spaced 91 cm apart. The center two rows served as treatment rows which received defoliant applications and were utilized for defoliation ratings and harvest, with the outer two rows of each plot serving as boarders. All other crop management practices followed state extension recommendations (Collins et al., 2015; Edmisten et al., 2015). Treatments included a factorial design with four cultivar options and two defoliation options. Cultivars included smooth leaf cultivars Deltapine 1028 B2RF (DP 1028) and Deltapine 1137 B2RF (DP 1137) (Monsanto 2012 Seed Resource Guide, 2011; Monsanto Company, St. Louis, MO), the semi-smooth cultivar PhytoGen 499 WRF (PHY 499) (Phytogen Seed Varieties, 2013; Dow AgroSciences, Indianapolis, IN), and the hairy cultivar Stoneville 5288 B2F (ST 5288) (Bayer CropScience Variety Selector, 2013; Bayer CropScience, Research Triangle Park, NC). Leaf pubescence ratings for three of the four varieties used in this study were found to be reported in the Arkansas Cotton Variety Test (Bourland et al., 2012; Bourland et al., 2013). On a scale from one to nine, with one being a smooth leaf and nine being very hairy, the DP 1028 cultivar was rated at a 1.1 in the 2011 report (Bourland et al., 2012), while the PHY 499 cultivar was rated at 3.8 and the ST 5288 cultivar was rated at 5.9 (with a 6.9 rating in 2011) in the 2012 report (Bourland et al., 2013). Two defoliation treatments were included; one which targets defoliation of the crop using recommended rates (Whitaker and Collins, 2015) appropriate for prevailing temperatures (recommended defoliation treatment) and one which includes a higher rate of the same defoliants plus the addition of a herbicidal defoliant, or a desiccating treatment (aggressive defoliation treatment). The recommended treatment included 0.73 L ha^{-1} of tribufos (Folex, Amvac Chemical Corporation, Newport Beach, CA) for defoliation, 2.34 L ha^{-1} of ethephon (Prep, Bayer CropScience, Research Triangle Park, NC) for boll opening, and 0.23 L ha^{-1} of thidiazuron (Freefall, Nufarm Americas, Inc., Alsip, IL) for regrowth control. The aggressive treatment included 1.17 L $ha^{-1}$of tribufos, 2.34 L ha^{-1} of ethephon, 0.23 L ha^{-1} of thidiazuron, and 0.15 L ha^{-1} of pyraflufen ethyl (ET, Nichino America Inc., Wilmington, DE) a herbicidal defoliant/desiccant. Applications were made using a CO_2 pressurized backpack sprayer calibrated to deliver 140.3 L ha^{-1} at 4.8 km h^{-1}. Applications were made when plants had reached a maximum of four nodes above cracked boll, a recommended defoliation time (Collins et al., 2015).

Treatment results were evaluated at one, two, and three weeks after the defoliation treatments (WAT) were applied, for visual ratings of percent of open bolls, percent defoliation, percent desiccation, percent regrowth from the terminal of the plant (RG_T), and percent basal regrowth (RG_B). At the North Carolina location, basal and terminal regrowth were combined into one total regrowth (RG) rating. After the three intervals of visual inspections of defoliation were completed, cotton was harvested with a John Deere 9930 (John Deere, Moline, IL) two-row plot harvester equipped with bagging attachments for small-plot harvest. Seedcotton from all plots from all locations were weighed prior to ginning at the University of Georgia Micro Gin in Tifton, Georgia for determination of lint percentage and lint yield (Li et al., 2011). After ginning, approximately 230 g of lint from each plot were sent to the USDA Classing Office in Macon, Georgia for both classing and high volume instrumentation (HVI) measurements of fiber quality.

Data were analyzed as a split block design utilizing SAS 9.4 software (SAS Institute, Cary, NC) with the fixed effect of defoliation treatment as the main block factor and the random effect of cultivar as the subplot factor. Four replications of each treatment were included at each site year. Treatment means were separated by Fisher's Protected LSD at $\alpha \leq 0.05$.

Results and Discussion

Excessive rainfall prior to the harvest during 2014 in Tifton and the shorter season environment in North Carolina prevented pooling data results across locations. Thus, locations were analyzed and reported independently. Weekly weather data for all three locations from the application of defoliants until the final rating date is included in Table 6.1.

Tifton, GA 2013 Defoliation Ratings. The interaction of cultivar by defoliation was significant at three WAT (Table 6.2), where the percent of open bolls was lower in the recommended defoliation of ST 5288 than all other cultivar and defoliation combinations with the exception of aggressive defoliation on DP 1137 (Fig. 6.1). It is unclear why percent of open bolls would be impacted in one specific cultivar, but the actual difference in the range between all treatments (98.5 to 100% open bolls) does not reflect any biological or applied difference between the treatments. Cultivar had a significant effect on open bolls, defoliation, and desiccation at one WAT and on open bolls at two WAT across both defoliation treatments (Table 6.2). However, the differences in cultivar did not follow the leaf pubescence categories, as PHY 499, a semi-smooth cultivar, resulted in greater percentages of open bolls at one and two WAT, defoliation at one WAT, and lower desiccation at one WAT than the smooth leaf cultivar, DP 1137, as well as the hairy leaf cultivar, ST 5288 (Table 6.3). A significant difference between the smooth leaf cultivars occurred at two WAT, when the percentage of open bolls was greater in DP 1028 than DP 1137 (Table 6.3). A greater percentage of open bolls at one WAT, as well as greater percent defoliation and reduced desiccation at one WAT, was observed in the smooth-leaf DP 1028 compared to the hairy leaf ST 5288 (Table 6.3). Cultivar had no effect on regrowth at any rating date. The differences observed in defoliation are likely due to differences between the cultivars independent of leaf pubescence levels, as no observations of leaf pubescence influencing open boll, defoliation, desiccation, or regrowth have been reported. Defoliation treatment had a significant effect on percent desiccation at one WAT, and percent defoliation and desiccation at two WAT, across all cultivars (Table 6.2). The aggressive defoliation method resulted in increased desiccation at one and two WAT, and reduced defoliation at two WAT (Table 6.3). Increased desiccation and reduced defoliation is to be expected, as the inclusion of the desiccants promotes rapid drying out of the leaf tissue, which results in a more rapid leaf death and prevents the formation of an abscission layer compared to the recommended defoliation treatment (Bovey and Miller, 1968; Brecke et al., 2001). There was no effect of regrowth due to defoliation strategy.

Tifton, GA 2014 Defoliation Ratings. Cultivar had no effect on open boll, defoliation, or RG_T percentages, but did impact desiccation at three WAT and RG_B at one and two WAT and across both defoliation treatments (Table 6.4). Cotton desiccation was lower in ST 5288 compared to all other cultivars, although the range of desiccation was only from 4 to 7% (Table 6.5). Greater RG_B was observed in the smooth-leaf cultivar DP 1028 than all other cultivars at one WAT (Table 6.5). By two WAT, RG_B was greatest in the two smooth-leaf cultivars. These differences are likely not attributed to leaf pubescence, and given the range of regrowth observed (3 to 6%), are not biologically significant from a crop management standpoint. When comparing cultivars pooled over defoliation treatments, desiccation differences were only noted at three WAT (Table 6.4). A significant effect of defoliation treatment, when pooled over cultivars occurred for cotton leaf defoliation at two WAT as well as for desiccation at all three rating dates (Table 6.4). Differences observed included 5% less defoliation and 6 to 10% more desiccation with the aggressive defoliation strategy (Table 6.5). There was no significant interaction between cultivars and defoliation strategy at this location.

Lewiston, NC 2014 Defoliation Ratings. The interaction of cultivar by defoliation treatment was significant for open bolls at one WAT (Table 6.6), where the aggressive defoliation treatment on DP 1137 resulted in a lower percentage of open bolls than the recommended defoliation treatment on DP 1028 and ST 5288 regardless of defoliation treatment (Fig. 6.2). The open boll percentages between all treatments ranged from 95 to 98%, thus it is unlikely that this difference is biologically significant and would impact crop management. Cultivar had a significant effect on both defoliation and desiccation at one and two WAT (Table 6.6). The separation in both defoliation and desiccation was at most 4% among cultivars (Table 6.6). At one WAT, defoliation in the hairy-leaf ST 5288 was significantly lower than the smooth-leaf DP 1028, while greater desiccation was observed in the hairy-leaf ST 5288 than in the two smooth leaf cultivars, DP 1028 and DP 1137 (Table 6.7). At two WAT, defoliation in PHY 499 and ST 5288 was lower than the two smooth leaf cultivars, while desiccation ratings in the two cultivars with greater levels of leaf pubescence were higher than DP 1137. The main effect of defoliation treatment did not have a significant effect on any of the parameters measured by defoliation ratings at Lewiston, NC 2014.

Over all locations, when significant differences were present, the aggressive defoliation treatment resulted in reduced defoliation and increased desiccation. This is expected, as defoliation methods that desiccate plant tissues often lead to sticking of desiccated leaves to the plant as the abscission layer that is necessary for proper defoliation is not formed (Bovey and Miller, 1968; Brecke et al., 2001). At the two Georgia site years, there was no observed pattern between leaf pubescence characteristics and defoliation practices for open bolls, defoliation, and desiccation percentages. At Lewiston, NC 2014, defoliation ratings were lower in the two cultivars with higher leaf pubescence ratings, PHY 499 and ST 5288, when significant differences were present. Similar results were present for desiccation ratings where the cultivar with the highest leaf pubescence rating, ST 5288, had increased percentages of desiccation at one and two WAT.

Lint Percentage and Yield. Cultivar had a significant effect on lint percentage at Tifton, GA 2013 and 2014, and on lint yield at Tifton, GA 2013 (Table 6.8). Lint percentage is typically dependent on specific cultivar characteristics, primarily seed size (Miller and Rawlings, 1967), which is most likely the primary cause behind the differences in lint percentage observed among the cultivars. A greater lint percentage was present in cultivars PHY 499 and DP 1028 compared to DP 1137 and ST 5288 at Tifton, GA 2013, while lint yield at this site year was higher in DP 1028 and PHY 499 than ST 5288 (Table 6.9). At Tifton, GA 2014 a significant difference in lint percentage was observed between all cultivars, with the highest and lowest lint percentage present in PHY 499 and ST 5288, respectively.

While lint percentage differences were significant among cultivars at Tifton, GA 2013 and 2014, the range was relatively small, with 2.1 and 3.1 percent difference among the highest and lowest cultivars in 2013 and 2014, respectively. The only instance in which cultivar had a significant effect on lint yield (Tifton, GA 2013), all cultivars yielded in excess of 2,000 kg ha^{-1} of lint, with a difference of 198.75 kg ha^{-1} of lint between the highest and lowest yielding cultivars (Table 6.9).

Fiber Quality. Previous studies have reported that the timing of defoliation can impact fiber quality properties such as length, strength, micronaire, and uniformity (Brown and Hyer, 1956; Snipes and Baskin, 1994; Faircloth et al., 2004; Karademir et al., 2007), while the selection of defoliation products has no effect on these fiber properties (Snipes and Baskin, 1994; Larson et al., 2005). Thus, the significant effect of cultivar on fiber quality properties such as staple, micronaire, strength, HVI length, and uniformity are reflective of the inherent genetic differences associated with the cultivars evaluated, as defoliation was timed appropriately to avoid premature defoliation and was uniform across all locations. The goal of this study was to determine the effect of leaf pubescence characteristics of cultivars and defoliation practices on the fiber quality properties such as leaf grade, color characteristics, and trash, thus these parameters are the focus of the results. However, differences in additional fiber quality parameters, primarily resulting from genetic cultivar effects, are also included.

There was no defoliation by cultivar interaction or defoliation main effects observed for any fiber quality parameters across all locations (Table 6.10). In contrast, cultivar main effects were noted for nearly every fiber quality property measured. Significant effects on the reflectance (Rd) and yellowness (+B) of the lint were observed at Tifton, GA 2014 and Lewiston, NC 2014. Although differences were noted, all results fell into the middling or strict low middling Rd categories, with the exception of one low middling sample from Tifton, GA 2013 and one strict middling sample from Lewiston, NC 2014 (data not shown). Additionally, all individual plot samples were in the white +B category (data not shown).

Cultivar also had a significant effect on HVI trash (a measure of percent surface area occupied by non-lint material in a sample, analyzed digitally) and leaf grade (measure of leaf content in a sample) (Cotton Inc., 2013). The cultivar with the highest leaf pubescence rating in the study, ST 5288, had an increased leaf grade and HVI trash values above all other cultivars at Tifton, GA 2014 and Lewiston, NC 2014 (Table 6.11). With the exception of HVI trash in

Tifton, GA 2014, PHY 499 resulted in greater leaf grade and HVI trash values than the two smooth leaf cultivars, though not as high as ST 5288.

Study results illustrate that open boll, defoliation, and desiccation percentages are influenced primarily by defoliation strategy or cultivar, with minimal interaction. This conclusion suggests leaf pubescence level has little influence on these factors; however, leaf pubescence characteristic of the cultivars was directly responsible for nearly all effects on fiber quality. Thus producers should be mindful of inherent leaf pubescence characteristics of cultivars and their potential influence on fiber quality and potential discounts or premiums for lint.

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Table 6.1. Weather data from Tifton, GA, 2013, 2014, and Lewiston, NC 2014 locations from defoliation to harvest.

Measurement		Tifton, GA 2013	
	1 WAT[z]	2 WAT	3 WAT
Min. Temperature (C)	19.7	15.7	17.6
Max. Temperature (C)	26.8	25.7	28.0
Average Temperature (C)	22.7	20.3	22.0
Rainfall (cm)	3.5	0.0	1.0
		Tifton, GA 2014	
	1 WAT	2 WAT	3 WAT
Min. Temperature (C)	21.6	22.2	18.8
Max. Temperature (C)	31.3	32.5	29.7
Average Temperature (C)	25.0	25.9	23.4
Rainfall (cm)	8.6	1.9	1.2
		Lewiston, NC 2014	
	1 WAT	2 WAT	3 WAT
Min. Temperature (C)	12.9	8.6	5.9
Max. Temperature (C)	24.6	20.3	23.8
Average Temperature (C)	18.3	14.0	14.5
Rainfall (cm)	2.6	0.1	0.0

[z] Temperature averages and rainfall totals for each week after treatment (WAT) defoliation ratings were conducted.

Table 6.2. Anova results (p-values) for cotton defoliation evaluations in Tifton, GA 2013. Factors include four cultivars, two defoliation treatments, and the interaction of cultivar by defoliation.

Factors	df	Open Bolls[Z]	Defoliation	Desiccation	RG_T	RG_B
1 WAT						
Cultivar[Y]	3	0.0062	0.0028	0.0296	N/A[W]	N/A
Defoliation[X]	1	0.5632	0.7471	0.0328	N/A	N/A
Cultivar*Defoliation	3	0.8607	0.6088	0.8947	N/A	N/A
2 WAT						
Cultivar	3	0.0157	0.0616	0.0594	0.4155	0.1097
Defoliation	1	0.3176	0.0326	0.0346	0.3910	1.0000
Cultivar*Defoliation	3	0.0864	0.9282	0.9132	0.4155	0.7002
3 WAT						
Cultivar	3	0.0959	0.0523	0.0523	0.7826	0.3080
Defoliation	1	0.4558	0.0754	0.0754	0.2152	0.6497
Cultivar*Defoliation	3	0.0205	0.624	0.624	0.7826	0.8062

[Z] Measurements include the percent of open bolls, defoliation, desiccation, terminal regrowth (RG_T), and basal regrowth (RG_B).

[Y] Cultivars include PhytoGen 499 WRF (PHY 499), Deltapine 1028 B2RF DP 1028), Deltapine 1137 B2RF (DP 1137), and Stoneville 5288 B2F (ST 5288).

[X] Defoliation treatments included a recommended defoliant mix (0.73 L ha^{-1} of tribufos, 2.34 L ha^{-1} of ethephon, and 0.23 L ha^{-1} of thidiazuron) and an aggressive (1.17 L ha^{-1} of tribufos, 2.34 L ha^{-1} of ethephon, 0.23 L ha^{-1} of thidiazuron, and 0.15 L ha^{-1} of pyraflufen ethyl) defoliant mix.

[W] No terminal or basal regrowth was recorded in any plots at 7 DAT.

Table 6.3. Main effect means for cultivar and defoliation treatment evaluations in Tifton, GA 2013.

Factor	Open Bolls[Z] (%)	Defoliation (%)	Desiccation (%)	RG_T (%)	RG_B (%)
1 WAT					
Cultivar [Y]					
PHY 499	92	89	8	0	0
DP 1028	90	81	9	0	0
DP 1137	86	73	12	0	0
ST 5288	84	66	15	0	0
$PLSD_{0.05}$	5	11	5	NS[W]	NS
Defoliation[X]					
Light	89	78	8	0	0
Aggressive	88	76	14	0	0
$PLSD_{0.05}$	NS	NS	5	NS	NS
2 WAT					
Cultivar					
PHY 499	100	94	6	0	0
DP 1028	98	92	8	0	1
DP 1137	98	90	10	0	0
ST 5288	97	88	12	0	0
$PLSD_{0.05}$	2	NS	NS	NS	NS
Defoliation					
Light	98	94	6	0	0
Aggressive	99	88	12	0	0
$PLSD_{0.05}$	NS	5	5	NS	NS
3 WAT					
Cultivar					
PHY 499	100	97	3	0	4
DP 1028	100	97	3	0	7
DP 1137	100	95	5	0	4
ST 5288	99	93	8	0	5
$PLSD_{0.05}$	NS	NS	NS	NS	NS
Defoliation					
Light	100	98	3	0	5
Aggressive	100	94	6	0	5
$PLSD_{0.05}$	NS	NS	NS	NS	NS

[Z] Parameters include percentages of open bolls, defoliation, desiccation, terminal regrowth (RG_T), and basal regrowth (RG_B).

[Y] Cultivars include PhytoGen 499 WRF (PHY 499), Deltapine 1028 B2RF DP 1028), Deltapine 1137 B2RF (DP 1137), and Stoneville 5288 B2F (ST 5288).

[X] Recommended defoliation treatment applied 0.73 L ha^{-1} of tribufos, 2.34 L ha^{-1} of ethephon, and 0.23 L ha^{-1} of thidiazuron. The aggressive treatment applied 1.17 L ha^{-1} of tribufos, 2.34 L ha^{-1} of ethephon, 0.23 L ha^{-1} of thidiazuron, and 0.15 L ha^{-1} of pyraflufen ethyl.

[W] No significant difference.

Table 6.4. Anova results (p-values) for cotton defoliation evaluations in Tifton, GA 2014. Factors include four cultivars, two defoliation treatments, and the interaction of cultivar by defoliation.

Factors	df	Open Bolls[Z]	Defoliation	Desiccation	RG_T	RG_B
1 WAT						
Cultivar[Y]	3	0.2393	0.7093	0.9770	0.0536	0.0260
Defoliation[X]	1	0.3368	0.9377	0.0435	0.4444	0.6376
Cultivar*Defoliation	3	0.2894	0.3130	0.8732	0.0992	0.8443
2 WAT						
Cultivar	3	0.3819	0.9909	0.2098	0.6765	0.0277
Defoliation	1	0.1705	0.0298	0.0081	0.8839	0.2619
Cultivar*Defoliation	3	0.7770	0.2359	0.2923	0.4265	0.3370
3 WAT						
Cultivar	3	N/A[W]	0.2343	0.0095	0.1181	0.1362
Defoliation	1	N/A	0.1272	0.0066	0.2773	0.4152
Cultivar*Defoliation	3	N/A	0.5068	0.3182	0.0781	0.4141

[Z] Measurements include the percent of open bolls, defoliation, desiccation, terminal regrowth (RG_T), and basal regrowth (RG_B).

[Y] Cultivars include PhytoGen 499 WRF (PHY 499), Deltapine 1028 B2RF DP 1028), Deltapine 1137 B2RF (DP 1137), and Stoneville 5288 B2F (ST 5288).

[X] Defoliation treatments included a recommended defoliant mix (0.73 L ha^{-1} of tribufos, 2.34 L ha^{-1} of ethephon, and 0.23 L ha^{-1} of thidiazuron) and an aggressive (1.17 L ha^{-1} of tribufos, 2.34 L ha^{-1} of ethephon, 0.23 L ha^{-1} of thidiazuron, and 0.15 L ha^{-1} of pyraflufen ethyl) defoliant mix.

[W] All plots had reached 100% open bolls at 3 WAT.

Table 6.5. Main effect means for cultivar and defoliation treatment evaluations in Tifton, GA 2014.

Factor	Open Bolls[Z] (%)	Defoliation (%)	Desiccation (%)	RG_T (%)	RG_B (%)
1 WAT					
Cultivar[Y]					
PHY 499	97	77	14	0	0
DP 1028	94	73	15	1	0.5
DP 1137	94	74	15	0	0
ST 5288	95	73	15	0	0.13
$PLSD_{0.05}$	NS[W]	NS	NS	NS	0.36
Defoliation[X]					
Light	94	74	10	0	0
Aggressive	96	74	20	0	0
$PLSD_{0.05}$	NS	NS	10	NS	NS
2 WAT					
Cultivar					
PHY 499	100	90	8	0	4
DP 1028	99	89	11	1	6
DP 1137	99	89	10	0	6
ST 5288	99	90	8	1	3
$PLSD_{0.05}$	NS	NS	NS	NS	2
Defoliation					
Light	99	92	6	0	4
Aggressive	100	87	12	0	6
$PLSD_{0.05}$	NS	4	3	NS	NS
3 WAT					
Cultivar					
PHY 499	100	94	6	1	11
DP 1028	100	93	7	2	12
DP 1137	100	83	7	1	13
ST 5288	100	95	4	1	9
$PLSD_{0.05}$	NS	NS	2	NS	NS
Defoliation					
Light	100	96	3	1	10
Aggressive	100	87	9	1	13
$PLSD_{0.05}$	NS	NS	3	NS	NS

[Z] Parameters include percentages of open bolls, defoliation, desiccation, terminal regrowth (RG_T), and basal regrowth (RG_B).

[Y] Cultivars include PhytoGen 499 WRF (PHY 499), Deltapine 1028 B2RF DP 1028), Deltapine 1137 B2RF (DP 1137), and Stoneville 5288 B2F (ST 5288).

[X] Recommended defoliation treatment applied 0.73 L ha^{-1} of tribufos, 2.34 L ha^{-1} of ethephon, and 0.23 L ha^{-1} of thidiazuron. The aggressive treatment applied 1.17 L ha^{-1} of tribufos, 2.34 L ha^{-1} of ethephon, 0.23 L ha^{-1} of thidiazuron, and 0.15 L ha^{-1} of pyraflufen ethyl.

[W] No significant difference.

Table 6.6. Anova results (p-values) for cotton defoliation evaluations in Lewiston, NC 2014. Factors include four cultivars, two defoliation treatments, and the interaction of cultivar by defoliation.

Factors	df	Open Bolls[Z]	Defoliation	Desiccation	RG
1 WAT					
Cultivar[Y]	3	0.0760	0.0420	0.0289	N/A[W]
Defoliation[X]	1	0.4338	0.1273	0.0563	N/A
Cultivar*Defoliation	3	0.0399	0.7197	0.4362	N/A
2 WAT					
Cultivar	3	0.2642	0.0096	0.0475	N/A
Defoliation	1	0.1027	0.0685	0.0675	N/A
Cultivar*Defoliation	3	0.3381	0.7179	0.7654	N/A
3 WAT					
Cultivar	3	N/A[V]	0.7708	0.7708	N/A
Defoliation	1	N/A	0.2113	0.2113	N/A
Cultivar*Defoliation	3	N/A	0.3308	0.3308	N/A

[Z] Measurements include the percent of open bolls (OB), defoliation (DEF), desiccation (DES), and regrowth (RG).

[Y] Cultivars include PhytoGen 499 WRF (PHY 499), Deltapine 1028 B2RF DP 1028), Deltapine 1137 B2RF (DP 1137), and Stoneville 5288 B2F (ST 5288).

[X] Defoliation treatments included a recommended defoliant mix (0.73 L ha^{-1} of tribufos, 2.34 L ha^{-1} of ethephon, and 0.23 L ha^{-1} of thidiazuron) and an aggressive defoliant mix (1.17 L ha^{-1} of tribufos, 2.34 L ha^{-1} of ethephon, 0.23 L ha^{-1} of thidiazuron, and 0.15 L ha^{-1} of pyraflufen ethyl).

[W] No regrowth was recorded at any evaluation date.

[V] All plots had reached 100% open bolls at 3 WAT.

Table 6.7. Main effect means for cultivar and defoliation treatment evaluations in Lewiston, NC 2014.

	Open Bolls[Z] (%)	Defoliation (%)	Desiccation (%)	RG (%)
1 WAT				
Cultivar[Y]				
PHY 499	97	95	3	0
DP 1028	97	97	2	0
DP 1137	95	96	3	0
ST 5288	97	94	5	0
$PLSD_{0.05}$	NS[W]	2	2	NS
Defoliation[X]				
Light	97	96	2	0
Aggressive	96	95	4	0
$PLSD_{0.05}$	NS	NS	NS	NS
2 WAT				
Cultivar				
PHY 499	98.88	94	6	0
DP 1028	99	97	3	0
DP 1137	98.63	98	2	0
ST 5288	99	94	6	0
$PLSD_{0.05}$	0.44	3	3	NS
Defoliation				
Light	99.06	97	2	0
Aggressive	98.69	94	6	0
$PLSD_{0.05}$	NS	NS	NS	NS
3 WAT				
Cultivar				
PHY 499	100	97	3	0
DP 1028	100	98	2	0
DP 1137	100	97	3	0
ST 5288	100	97	3	0
$PLSD_{0.05}$	NS	NS	NS	NS
Defoliation				
Light	100	98	2	0
Aggressive	100	96	4	0
$PLSD_{0.05}$	NS	NS	NS	NS

[Z] Parameters include percentages of open bolls, defoliation, desiccation, and regrowth (RG).

[Y] Cultivars include PhytoGen 499 WRF (PHY 499), Deltapine 1028 B2RF DP 1028), Deltapine 1137 B2RF (DP 1137), and Stoneville 5288 B2F (ST 5288).

[X] Recommended defoliation treatment applied 0.73 L ha^{-1} of tribufos, 2.34 L ha^{-1} of ethephon, and 0.23 L ha^{-1} of thidiazuron. The aggressive treatment applied 1.17 L ha^{-1} of tribufos, 2.34 L ha^{-1} of ethephon, 0.23 L ha^{-1} of thidiazuron, and 0.15 L ha^{-1} of pyraflufen ethyl.

[W] No significant difference.

Table 6.8. Anova results (p-values) for cotton yield parameters in Tifton, GA 2013 and 2014, and Lewiston, NC 2014. Factors include four cultivars, two defoliation treatments, and the interaction of cultivar by defoliation.

Factors	df	Seedcotton Yield[Z]	Lint Percentage	Lint Yield
Georgia 2013				
Cultivar[Y]	3	0.3185	0.0077	0.0336
Defoliation[X]	1	0.2378	0.5644	0.3071
Cultivar*Defoliation	3	0.6461	0.9850	0.6597
Georgia 2014				
Cultivar	3	0.0513	<.0001	0.1131
Defoliation	1	0.4915	0.7381	0.4415
Cultivar*Defoliation	3	0.9750	0.8554	0.9688
North Carolina 2014				
Cultivar	3	0.6461	0.3457	0.2100
Defoliation	1	0.9013	0.3674	0.4471
Cultivar*Defoliation	3	0.4478	0.5025	0.5933

[Z] Measurements include the seedcotton yield, lint percentage, and lint yield.

[Y] Cultivars include PhytoGen 499 WRF (PHY 499), Deltapine 1028 B2RF DP 1028), Deltapine 1137 B2RF (DP 1137), and Stoneville 5288 B2F (ST 5288).

[X] Defoliation treatments included a recommended defoliant mix (0.73 L ha^{-1} of tribufos, 2.34 L ha^{-1} of ethephon, and 0.23 L ha^{-1} of thidiazuron) and an aggressive defoliant mix (1.17 L ha^{-1} of tribufos, 2.34 L ha^{-1} of ethephon, 0.23 L ha^{-1} of thidiazuron, and 0.15 L ha^{-1} of pyraflufen ethyl).

Table 6.9. Main effect means for cultivar on cotton yield parameters in Tifton, GA 2013 and 2014, and Lewiston, NC 2014. Cultivar means are pooled over defoliation treatments.

Cultivar[Z]	Seedcotton Yield (kg/ha)[Y]	Lint Percentage (%)	Lint Yield (kg/ha)
Tifton, GA 2013			
PHY 499	4755.71	41.3	2201.26
DP 1028	4663.02	40.8	2133.05
DP 1137	4765.14	39.2	2090.20
ST 5288	4522.73	39.5	2002.51
PLSD$_{0.05}$	NS[X]	1.3	130.11
Tifton, GA 2014			
PHY 499	2964.50	43.2	1434.46
DP 1028	2622.68	42.2	1239.79
DP 1137	2920.13	41.1	1344.30
ST 5288	3418.6	40.1	1530.55
PLSD$_{0.05}$	NS	1.0	NS
Lewiston, NC 2014			
PHY 499	1601.70	44.2	794.46
DP 1028	1537.27	44.1	760.32
DP 1137	1500.49	37.9	630.95
ST 5288	1521.06	42.3	719.97
PLSD$_{0.05}$	NS	NS	NS

[Z] Cultivars include PhytoGen 499 WRF (PHY 499), Deltapine 1028 B2RF DP 1028), Deltapine 1137 B2RF (DP 1137), and Stoneville 5288 B2F (ST 5288).

[Y] Parameters include seedcotton yield, lint percentage, and lint yield.

[X] No significant difference.

Table 6.10. Anova results (p-values) from lint quality parameters from Tifton, GA 2013 and 2014, and Lewiston, NC 2014. Factors include cultivar, defoliation treatment, and the interaction of cultivar by defoliation (Cult*Defol).

Factors	df	Staple[Z]	Mic	Strength	Leaf Grade	Rd	+B	HVI trash	HVI length	Uniformity
Georgia 2013										
Cultivar[Y]	3	0.0249	0.0014	0.0010	0.5302	0.0852	0.8043	0.6793	0.0363	0.4561
Defoliation[X]	1	0.1817	0.0679	0.8185	0.3966	0.7952	0.7244	0.7827	0.3659	0.4829
Cultivar*Defoliation	3	0.7272	0.1564	0.6689	0.9738	0.4277	0.8348	0.9141	0.8852	0.9993
Georgia 2014										
Cultivar	3	0.0004	<.0001	0.0021	<.0001	0.0356	<.0001	<.0001	<.0001	0.0114
Defoliation	1	0.5456	0.1470	0.7099	0.8116	0.7483	0.8601	0.3132	0.6209	0.4446
Cultivar*Defoliation	3	0.1541	0.9798	0.8249	0.5581	0.6976	0.5180	0.9051	0.1777	0.5882
North Carolina 2014										
Cultivar	3	0.0895	0.0221	<.0001	<.0001	<.0001	<.0001	<.0001	0.0436	<.0001
Defoliation	1	0.3563	0.9675	0.2587	0.6069	0.2255	0.4729	0.8760	0.1768	0.6736
Cultivar*Defoliation	3	0.6871	0.5513	0.5721	0.6194	0.3933	0.2501	0.8993	0.2137	0.0411

[Z] Measurements include color grade, staple, micronaire (Mic), strength, leaf grade, reflectance (Rd), yellowness (+B), HVI trash, HVI length, and uniformity.

[Y] Cultivars include PhytoGen 499 WRF (PHY 499), Deltapine 1028 B2RF DP 1028), Deltapine 1137 B2RF (DP 1137), and Stoneville 5288 B2F (ST 5288).

[X] Defoliation treatments included a recommended defoliant mix (0.73 L ha^{-1} of tribufos, 2.34 L ha^{-1} of ethephon, and 0.23 L ha^{-1} of thidiazuron) and an aggressive defoliant mix (1.17 L ha^{-1} of tribufos, 2.34 L ha^{-1} of ethephon, 0.23 L ha^{-1} of thidiazuron, and 0.15 L ha^{-1} of pyraflufen ethyl).

Table 6.11. Main effect means of cultivar on of fiber quality characteristics from Tifton, GA 2013 and 2014, and Lewiston, NC 2014. Cultivar means are pooled over defoliation treatments.

Cultivar[Z]	Staple[Y] (32nds of an inch)	Mic	Strength (g/tex)	Leaf Grade	Rd	+B	HVI Trash (% area)	HVI Length (inches)	Uniformity (%)
Tifton, GA 2013									
PHY 499	36.25	4.76	30.94	3.88	74.61	7.84	0.613	1.13	83.26
DP 1028	37	4.59	28.51	3.13	75.94	7.95	0.44	1.15	82.91
DP 1137	36.63	4.49	28.89	4	76.29	7.71	0.64	1.14	82.59
ST 5288	37	4.41	28.923	3.63	76.51	7.73	0.56	1.15	82.79
$\text{PLSD}_{0.05}$	0.536	0.159	1.11	NS[X]	NS	NS	NS	0.02	NS
Tifton, GA 2014									
PHY 499	35.63	4.89	31.78	3	75.05	8.85	0.38	1.11	82.95
DP 1028	36.75	4.75	29.80	2.17	76.29	8.93	0.24	1.14	83.4
DP 1137	37	4.6	29.89	2.13	76.25	8.73	0.28	1.15	82.85
ST 5288	36.38	4.91	29.44	3.88	75.51	7.78	0.6	1.13	81.94
$\text{PLSD}_{0.05}$	0.55	0.1	1.18	0.48	0.95	0.26	0.1	0.01	0.86
Lewiston, NC 2014									
PHY 499	36.5	4.85	31.88	3.25	76.79	7.76	0.49	1.14	84.41
DP 1028	36.88	4.79	28.86	1.57	79.43	7.89	0.17	1.15	84.24
DP 1137	36.75	4.83	28.923	2	79.58	7.76	0.21	1.14	84.31
ST 5288	36.25	4.61	29.34	4	78.96	7.08	0.63	1.13	82.74
$\text{PLSD}_{0.05}$	NS	0.16	1.07	0.45	0.97	0.19	0.14	0.01	0.57

[Z] Cultivars include PhytoGen 499 WRF (PHY 499), Deltapine 1028 B2RF DP 1028), Deltapine 1137 B2RF (DP 1137), and Stoneville 5288 B2F (ST 5288).

[Y] Measurements include color grade, staple, micronaire (Mic), strength, leaf grade, reflectance (Rd), yellowness (+B), HVI trash, HVI length, and uniformity.

[X] No significant difference.

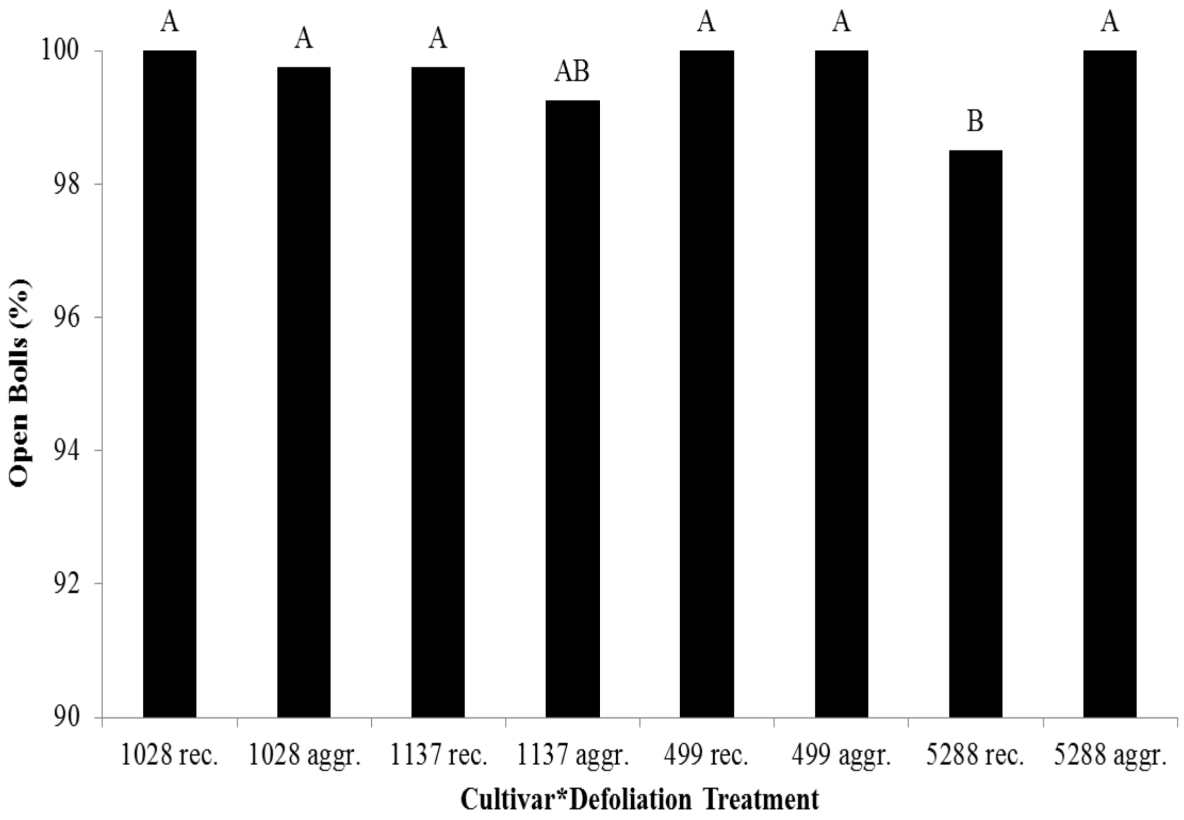

Figure 6.1. Percent open bolls as influenced by the interaction of cultivars DP 1028 (1028), DP 1137 (1137), PHY 499 (499), and ST 5288 (5288) and the recommended (rec.) and aggressive (aggr.) defoliation treatments at three WAT in Tifton, GA 2013.

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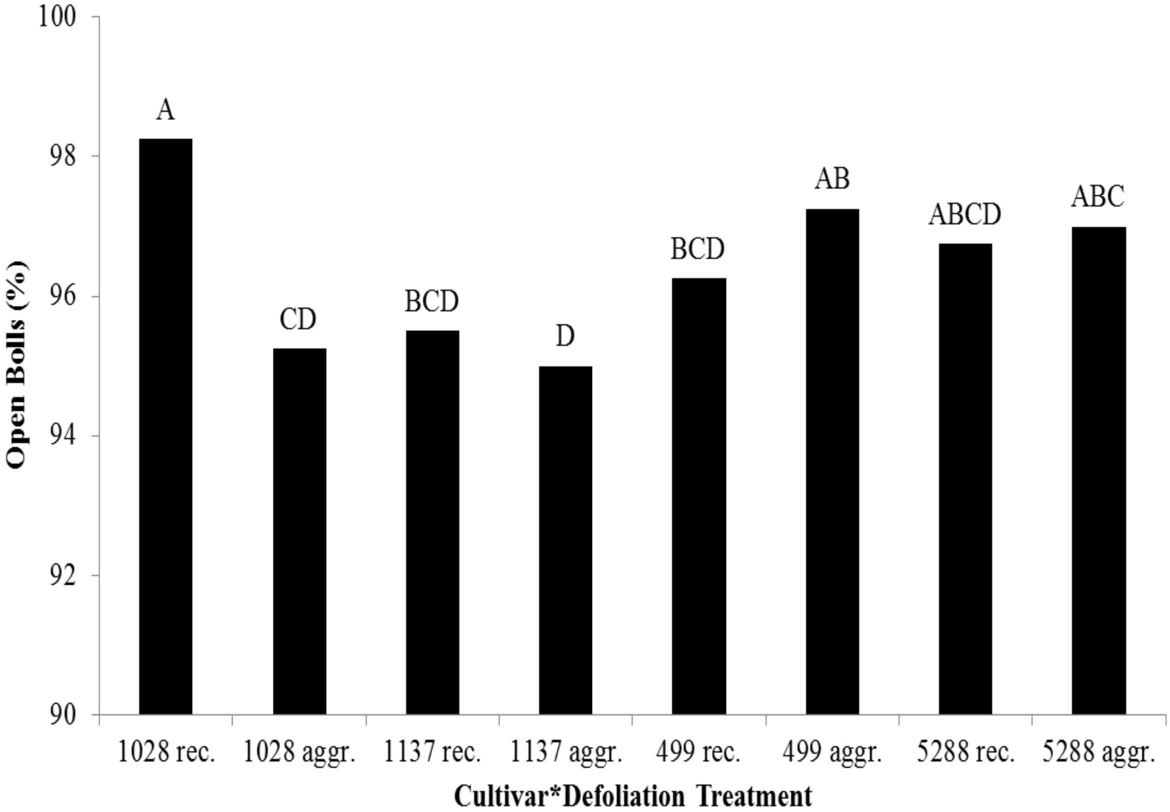

Figure 6.2. Percent open bolls as influenced by the interaction of cultivars DP 1028 (1028), DP 1137 (1137), PHY 499 (499), and ST 5288 (5288) and the recommended (rec.) and aggressive (aggr.) defoliation treatments at one WAT at the Lewiston, NC 2014.

CHAPTER 7

SUMMARY AND CONCLUSIONS

Impact of Sub-Lethal 2,4-D Rates on Cotton. New technologies enabling post-emergence applications of 2,4-D on cotton contribute an additional mode-of-action for weed control, while also creating an interface between cultivars with and without resistance to the herbicide. As one of the most sensitive crop species to 2,4-D (Robinson and Fox, 1978; Bayley et al., 1992; Lyon et al., 1993), cotton injury due to off-target movement or misapplication of the herbicide has the potential to result in significant yield loss. Research has shown a detrimental yield effect from exposure of cotton at early growth stages to sub-lethal rates of 2,4-D (Sciumbato et al., 2004; Marple et al., 2007, 2008; Everitt and Keeling, 2009; Johnson et al., 2012a; Egan et al., 2014). With the ability to apply the herbicide to cotton directly, the opportunity for incidences of off-target movement onto adjacent cotton occurring at a wide range of growth stages will exist. Determining the sensitivity of these various growth stages, as well as evaluating methods for predicting injury will be critical in determining appropriate management practices to be utilized when these incidents occur. The use of fluorescence measurements has been successful in the past at detecting exposure of non-crop species to an array of PS I and PS II inhibiting herbicides (Haynes et al., 2000; Ralph, 2000; Eullaffroy and Vernet, 2003), though this method is untested in detecting synthetic auxin herbicides in crop plants, cotton in particular.

The results of this study revealed that growth stages near the early bloom period are the most sensitive to sub-lethal rates of 2,4-D in regards to yield loss, primarily through a reduction in boll number. This is in contrast to previous work which reported early, vegetative growth stages being the most sensitive to sub-lethal rates of the herbicide (Egan et al., 2014). Visual evaluations of injury symptoms did not reflect the yield loss sustained at the most sensitive growth stages, likely due to the lack of vegetative symptoms typically observed from 2,4-D exposure. Measurements of chlorophyll *a* fluorescence parameters produced no value in regards to predicting yield loss or even the presence of 2,4-D exposure on cotton. There was no impact on fluorescence properties of cotton due to sub-lethal 2,4-D applications, even when severe yield loss resulted. Moving forward, additional research will be needed to determine and develop successful methods for quantifying injury symptoms to reproductive structures so that yield loss predictions can be made and recommendations for mitigating management practices can be refined.

Agronomic and Physiological Benefits of Conservation Tillage with High Biomass Rolled Rye Cover Crop. As concerns and regulations increase on the subject of agricultural water use, implementation of management practices that allow for more efficient use of both irrigation and unpredictable rainfall will benefit producers of large acreage crops such as cotton. The detrimental effects of water deficit on cotton yield are well documented (Gerik et al., 1996; Pringle and Martin, 2003; Pettigrew, 2004; Whitaker et al., 2008; Gwathmey et al., 2011), as are the effects on physiological process governing gas exchange, photosynthesis (Medrano et al., 2002; Flexas et al., 2002; Snider et al., 2014), and plant water status through leaf water potential (McMichael et al., 1973; Ackerson et al., 1977a; Ackerson et al., 1977b; Ackerson and Hebert, 1981; Radin, 1984; Turner et al., 1986; Snider et al., 2014; Chastain et al., 2014). Research on the use of cover crops in agriculture has revealed benefits to soil moisture (Blevins et al., 1971; Gantzer and Blake, 1978; Mills et al., 1988; Dao, 1993; Daniel et al., 1999), crop growth (Bauer and Busscher, 1996; Bauer et al., 2010), and yield (Bordovsky et al., 1994; Bauer and Busscher, 1996; Raper et al., 2000; Schomberg et al., 2006; Wiatrak et al., 2006; Bauer et al., 2010). Evaluating the potential water saving benefits of the use of cover crops, and any resulting benefits to crop growth and yield could provide producers with a management strategy to combat water deficits resulting from episodic drought periods and allow for more efficient use of irrigation. Investigating the effect of a conservation tillage system utilizing a cover crop on the underlying physiological processes impacted by crop water status would provide understanding on the crop response to differing tillage practices.

Due to rainfall that occurred throughout both years of the study, little benefits were observed through the use of conservation tillage utilizing a high biomass rolled rye cover crop compared to conventional tillage. This was particularly true for the 2013 season, during which multiple events of excessive rainfall occurred, resulting in reduced growth and yield in the conservation tillage treatment. Results from varying irrigation agreed with previous research, as cotton growth, development, and yield generally increased with increasing water supply, although in several instances no differences were present between the irrigation treatments that applied the two greatest amounts. Similar to the agronomic portion of the study, little physiological benefit was observed due to conservation tillage compared to conventional tillage. However, irrigation did have an effect, as increased stomatal conductance and one instance of higher photosynthetic rates were observed when comparing fully irrigated to nonirrigated cotton. These instances coincided with increases in leaf water potential in the fully irrigated treatment, which is strongly linked to stomatal conductance (Radin, 1984; Turner et al., 1986; Snider et al., 2014), agreeing with previous findings. Past research that reported benefits to crop growth and yield resulting from cotton grown under conservation tillage compared to conventional were conducted in water deficit or drought conditions (Bauer et al., 2010). Such water-limiting conditions were not present during this study, thus the potential benefits, or even detriments, resulting from this conservation tillage system appear to be dependent on environmental conditions. Further work, conducted in more severe water deficit conditions than were present during the current study, would be needed to fully comprehend the benefits gained from this conservation tillage, high biomass rolled rye system.

Influence of Cultivar Leaf Pubescence and Defoliation Strategy on Cotton Fiber Quality. Defoliation is often required for indeterminate crops like cotton, with profitability and quality maximized through appropriate defoliation practices. When performed properly, defoliation will decrease the amount of leaf or other plant material present in harvested seedcotton and ginned lint (Brecke et al., 2001; Valco and Snipes, 2001). There are two primary categories of harvest aides that serve as defoliants, or products that remove vegetation from cotton; defoliants and desiccants. Defoliants remove vegetation through the formation of an abscission layer at the base of the petiole when applied to mature, healthy leaves, resulting in the leaves falling off the plant (Cathey, 1986; Brecke et al., 2001). Application of desiccants, or herbicidal defoliants, leads to rapid death of leaves preventing the formation of an abscission layer (Stahler, 1953; McMeans et al., 1966; Bovey and Miller, 1968; Brecke et al., 2001; Shaw, 2002). The use of desiccants often results in dead leaves remaining on the plant at harvest, increasing the amount of leaves present in harvested seedcotton (Shaw, 2002). Leaf pubescence level of cultivars is another factor that can contribute to increases in leaf or plant material in lint. Cultivars with greater pubescence, or “hairy leaf cultivars” typically have greater leaf or trash content due to the pubescent plant material becoming entangled in the lint of open bolls upon defoliation (Ramey, 1962; Smith, 1964; Wanjura et al., 1976; Novick et al., 1991). Determining the influence of leaf pubescence, as well as defoliation strategy, on fiber quality, can provide producers with key information regarding cultivar selection and defoliation strategy.

Overall, there was minimal impact of defoliation strategy on cotton defoliation parameters across the three trial locations during the two year study. The desiccating defoliation strategy resulted in reduced defoliation and increased desiccation ratings, when significant. However, none of the differences would be considered biologically significant, or resulting in a difference in management practices due to defoliation strategy, as both defoliation strategies resulted in adequate cotton defoliation. Two smooth leaf and two “hairy” leaf cultivars were included, although when cultivar was significant for parameters used in defoliation ratings, there was no clear pattern observed. Specifically, smooth leaf cultivars were not in contrast to the hairy leaf cultivars when differences were present. For fiber quality measurements, the focus of this study was leaf grade (leaf material in ginned lint) and trash (non-leaf material in ginned lint). While defoliation strategy had no effect on these fiber quality measurements, cultivar was significant at two of the three locations. In these instances, the cultivars with higher levels of leaf pubescence resulted in increased leaf grade and trash compared to the smooth leaf cultivars. The lack of interaction between cultivar and defoliation strategy for fiber quality illustrates that leaf pubescence level of cultivars has a greater influence on the fiber quality parameters leaf grade and trash, thus producers should be mindful of cultivar characteristics, and potential quality discounts or premiums to maximize profitability.

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