ASSESSING GENOTYPIC AND SOIL MANAGEMENT RESOURCES FOR IMPROVING RESILIENCE IN SOUTHEASTERN UNITED STATES ROW CROP PRODUCTION SYSTEMS

By

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To my Mother and Father

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LIST OF ABBREVIATIONS

Akaike information criterion (AIC)	Estimator of the relative quality/fit of a statistical model. Lower number defines model as a relatively better estimator.
Analysis of variance (ANOVA)	Statistical models used to analyze the variance among group means for determining differences.
Cellulose acetate butyrate (CAB)	Material composition of mini-rhizotron tubes.
Coordinated adaptive phenotyping (CAP)	Term referring to the assessment of pertinent crop traits functioning in unison to provide a biological advantage in a particular cropping production system. Used in context to soil water acquisition and utilization in the south east production system in this dissertation.
Days after planting (DAP)	Number of days after crop planting/sowing.
Dehydration vulnerability scenario (DVS)	This term refers to general water-deficit conditions which are relevant to a particular production system as a result of climate, environmental conditions, and production management.
Economically optimum nitrogen rate (EONR)	Defined as the nitrogen rate that maximizes the value of crop produced at a given crop and fertilizer price.
Fischer's least significant difference (LSD)	A statistical method for determining confidence intervals between pairwise comparisons while controlling for a specified experiment-wise error rate.
General linear mixed model (GLIMMIX)	Statistical model used for computing an analysis of variance.
Genotype by environment interaction (GxE)	Infers that crop genotypic traits are responding differently due to variation in environmental conditions.
Genotype by environment by management interaction (GxExM)	Infers that crop genotypic traits are responding differently due to variation in environmental conditions and management decisions.
Genotype by management	Infers that crop genotypic traits are responding differently due to variation in management decisions.

Interaction (GxM)	
Genotype by year interaction (GxY)	Infers that crop genotypic traits are responding differently due to differences among growing seasons.
Harvest index (HI)	Term reefing to the proportion of harvestable crop biomass to total aboveground biomass.
Autoregressive (AR1)	Covariance matrix structure used in repeated measures analysis of variance to avoid violating independence assumption. Structure has homogenous variance and correlation among variances in time decline exponentially with distance.
Heterogeneous autoregressive (ARH1)	Covariance matrix structure used in repeated measures analysis of variance to avoid violating independence assumption. Structure has heterogeneous variance and correlation among variances in time decline exponentially with distance.
Nitrogen use efficiency (NUE)	Proportion of nitrogen in above-ground dry biomass to the total amount of nitrogen applied to the system.
Internal nitrogen use efficiency (iNUE)	Above-ground biomass produced per unit of tissue nitrogen.
Leaf area index (LAI)	Unit area of crop canopy per soil area.
New Mexico Valencia C (NMVC)	Valencia peanut genotype of *fastigiata* descent.
Plant available water (PAW)	The length of water available to a plant at a particular time period during the growing season. Estimated between the product of the effective root zone (length units) and the available water holding capacity of the soil.
Plant available water replacement (PAWR)	The percent of plant available water replacement from applying irrigation.
Plant science research and education unit (PSREU)	Research facility near Citra, FL where crop research was conducted.
Primed acclimation (PA)	Irrigation management practice of using regulated deficit irrigation during the predominantly vegetative growth stages of a crop to alter homeostasis for improving later season drought resiliency.
Regulated deficit	An irrigation practice designed to reduce the amount of water

irrigation (RDI)	application below the full water requirement for optimum plant growth.
Rain-fed (RF)	Irrigation treatment in where precipitation is the only form of water application.
Root length density (RLD)	Total longitudinal length of root segment per volume of soil.
Root system architecture (RSA)	Root parameters defined over the soil profile to make inference on root distribution through the soil.
Soil water depletion (SWD)	Measurement technique quantifying the amount of soil water depletion by examining the difference in volumetric soil water content from pre-dawn to pre-dusk.
Time domain reflectometry (TDR)	Measurement technique to quantify volumetric soil water content by assessing the reflection of an electromagnetic pulse.
Total root length (TRL)	Total longitudinal length of root segment per distance of soil.
Total root volume (TRV)	Total root volume per distance of soil calculated where $V=\Pi r^2*TRL$
Total surface area (TSA)	Total cross sectional root area per distance of soil where $CSA= 2\Pi r*TRL$
Total water received as a percentage of reference evapotranspiration (TWRPET)	Amount of total water (precipitation + irrigation) received in proportion to the amount of reference evapotranspiration defined by Penman-Monteith equation.
Transient chlorophyll fluorescence (OJIP)	Method for quantifying both physical and chemical energy transfer within the light reaction of photosynthesis.
Volumetric soil water content (VSWC)	Volume of water in proportion to the volume of soil.
Vapor pressure deficit (VPD)	The difference in water potential between the sub-stomatal cavity of a plant and the atmosphere.
Variable rate	Irrigation technology which allows for custom water application at

irrigation	specific location within a field.
100%	Refers to positive control irrigation treatment.
60%	Refer to season long reduction in irrigation application.
60%PA	Primed acclimation treatment consisting of a 40% reduction of the 100% treatment.

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Irrigation water and nitrogen (N) fertilizer are two of the most significant management inputs in southeast agronomic production systems for mitigating abiotic stress. These inputs not only influence production, but are a significant cost, both determining farmer profitability. Additionally, both inputs impact water consumption and quality. As the human population increases, so does the competition for quality freshwater between the residential and agricultural sectors. Research that assesses irrigation and N management optimization has the potential to positively influence these complex socio-economic and environmental issues.

One of the most challenging aspects of agronomic management is the complex interaction it has across production systems with varying crops, soil landscapes, and climatic conditions. Furthermore, stochastic yearly weather conditions often result in scenarios where management of water and N cannot meet crop demand resulting in yield loss due to abiotic stress. Fortunately, a large amount of genetic diversity exists within crop species possessing traits which can be utilized to mitigate yield loss under abiotic stress, and ultimately increasing long term food and fiber production.

Therefore, the proceeding chapters in this dissertation address both the optimization of water and N management, and evaluation of whole plant genotypic trait responses for determining crop adaptability to environment and soil hydrologic conditions. Chapter 1 of this dissertation will provide a general overview of the southern U.S. row crop production system with an emphasis on nitrogen and water management practices, providing the reader an introduction to the research chapters included in this dissertation. The research chapters will specifically address: (i) peanut genotypic pod yield adaptability to irrigation management across U.S. production regions; (ii) intrinsic above- and below-ground traits of disparate peanut genotypes for determining adaptability to soil hydrologic conditions; (iii) the coordination of traits which improve peanut soil water acquisition and utilization in the southeast irrigation production system; (iv) utilizing irrigation scheduling tools and in-season N management strategies to optimize cotton production in the southeast. The final conclusions chapter provides a brief synthesis of the major findings from this research.

CHAPTER 1
LITERATURE REVIEW

Introduction

Southern U.S. cropping systems: Two of the predominant rotational row crops in the southern United States (U.S.) are peanut (*Arachis hypogaea* L.) and upland cotton (*Gossypium hirsutum* L.).The southern U.S. can generally be delineated into specific regions with states of Florida, Georgia, Alabama, and Mississippi in the southeast; Virginia, North Carolina, and South Carolina in the east; and Texas, New Mexico, and Oklahoma in the southwest. These three regions (southeast, east, and southwest) accounted for an average of 73, 16, and 11% of the total area planted to peanut from 2012-2014, respectively (USDA-NASS, 2015). Cotton production in these three regions (southeast, east, and southwest) accounted for an average of 23, 9 and 68% of the total area planted in 2016, respectively (USDA-NASS, 2015).These regions also encompass extreme variation in disease pressure, temperature, humidity, rainfall patterns, and growing season duration. Two wide regional environmental characterizations can be made: 1) the southwestern production areas are characterized as having a semi-arid climate with low annual rainfall and high vapor pressure deficits (VPD); and 2) the southeast and eastern production areas are characterized as having humid conditions with high total annual precipitation and, in some years, high frequency of precipitation events. Although the humid southeastern and eastern regions typically receive more rainfall than the southwestern region, growing season rainfall patterns can be unpredictable and geographically isolated, which when combined with highly permeable soils in some areas, creates a need for supplemental irrigation. The potential for yield reductions across both regions due to large ranges of GxExM creates a need to

explore the responses of genetically diverse crop genotypes in an effort to increase yield potential across both environments, and the examination of management practices specific to each region for optimizing inputs to increase grower profitability and reduce detriments to the environment.

Peanut and cotton irrigation practices: One critically important management component that is prevalent across the southeast and southwest crop production systems is irrigation. Irrigated peanut acreage has recently accounted for 62 and 94% of the harvested peanut land area in the major peanut producing regions of the southeast and southwest, respectively (USDA-NASS, 2014). Irrigated cotton acreage has recently accounted for 46 and 59% of the harvested cotton land area in the major peanut producing regions of the southeast and southwest, respectively (USDA-NASS, 2014). This heavy reliance on irrigation across regions regardless of annual precipitation averages, shows the importance of supplemental water to combat the uncertainty in precipitation patterns that may decrease yield and impact grower profitability. Research in the southeastern U.S. has shown that overall yield increases are observed with irrigation; however obtaining a positive net return on irrigation cost can largely be dependent on peanut prices and crop rotation practices (Lamb et al., 1997; Lamb et al., 2007). Therefore, optimization of irrigation is critical in the southeast environment to ensure a positive net return on irrigation.

Crop Phenological Sensitivity to Water Stress: Peanut is an indeterminate annual crop, meaning that vegetative and reproductive stages overlap with one another. Developmental stages of peanut are classified as: (V_E) emergence; (V_o) open cotyledons; (V1) first tetrafoliate; (R1) beginning bloom; (R2) beginning peg; (R3)

beginning pod; (R4) full pod; (R5) beginning seed; (R6) full seed; (R7) beginning maturity; (R8) harvest maturity (Boote, 1982). Peanut water use is low after emergence, and increases linearly with increasing LAI until maximal water use, which generally occurs from R4-R7 (Stansell et al., 1976). The peak reproductive development stages from R4-R7 is when peanut has been documented to be most susceptible to yield loss from water deficit, and reports have documented that water deficit stress during peg and pod development (R2-R4) most greatly reduce pod yield (Stirling et al., 1989; Patel and Gangavani, 1990; Meisner 1991). However, the pre-flowering phase of peanut has been reported as a less sensitive time in phenological development to water deficit stress (Kulkarni et al., 1988; Patel and Golakiya, 1988; Rao et al., 1988).

Cotton is also an indeterminate annual crop where ranges in reproductive sensitivity to water deficit stress have been documented. General developmental stages of cotton are vegetative development from seedling emergence to first square (start of reproductive growth), first bloom, first developed boll, and first open boll (Oosterhuis, 1990). Sensitive developmental time periods when cotton is most susceptible to lint yield loss due to water deficit stress are from square initiation through boll development, with the most sensitive time period being the first half of boll development (Hake and Grimes, 2010). Additionally, large fluctuations in soil water conditions can cause abscission of reproductive fruits leading to lint yield losses (squares, flowers and bolls) (Hake and Grimes, 2010).

Use of Regulated Deficit Irrigation (RDI): Varying yield loss sensitivities to water deficit stress over the course of crop development have led to the examination of using developmental stage-based RDI. Specifically, vegetative RDI is the practice of

reducing water application below the full water requirement for optimum plant growth during vegetative growth when the crop is less sensitive to water deficit stress. This results in water savings with no yield reductions (Chai et al., 2016). Rowland et al. (2012) reported no decrease in peanut yield in one site year when plants were treated with early and mid-season irrigation deficit followed by no late season irrigation deficit when compared to season long no-deficit irrigation. Sustained peanut yields resulted in similar economic returns when peanuts were treated with a 50% irrigation reduction for the first 45 DAP, followed by no deficit irrigation for the remainder of the growing season when compared to no deficit irrigation for the entire growing season. Furthermore, the use of vegetative RDI in peanut has been demonstrated to increase root proliferation deeper in the soil which could possibly increase water deficit stress resilience (Rowland et al., 2012; Thangthon et al., 2016).

However, root distribution and pod yield responses have been reported to vary among peanut genotypes demonstrating variation in phenotypic plasticity (Jongrungklang et al., 2011). Furthermore, the phenotypic plasticity to increase root distribution deeper into the soil profile during vegetative water stress is likely heavily influenced by environmental conditions. This is of particular importance since success using vegetative RDI has been documented in arid environments (Rowland et al., 2012), or under controlled conditions with rainout shelters (Jongrungklang et al., 2011; Thangthon et al., 2016). In both scenarios, total water received is primarily through irrigation in contrast to peanut production in humid environments where stochastic precipitation is the primary water source, and water stress is more probable to be mild. Therefore, this previous work could be further advanced by examining both above- and

below-ground response of disparate peanut genotypes to various irrigation management strategies in the southeast humid environment for determining optimal irrigation strategies, and possibly more water deficit stress tolerant germplasm.

The use of vegetative RDI has been examined for cotton in the southeast production environment. Meeks et al (2017) reported no lint yield reductions when comparing pre-bloom irrigation scheduling thresholds of -70 and -100 kPa to -20 kPa. Yield reductions were reported in one of the drier years of the study when water was withheld after the pre-bloom period. These results indicate that reduction of water until mid-bloom is a viable water saving strategy that can increase grower profitability and agronomic water use efficiency. However, the use of soil water tension sensors used to schedule irrigation, as employed in this study, can be costly, and may not be feasible in some cotton production systems. Free ET-based soil water balance models have been developed for cotton (Vellidis et al., 2016) which provides site specific recommendations for irrigation scheduling. Providing information in regards to RDI management using this tool could be useful for growers to optimize irrigation management.

N requirement in cotton: Another major variable cost in cotton production as compared to peanut is nitrogen (N) fertilization, which is the predominate contributor of the overall fertilization cost of cotton and usually exceeds the irrigation costs (Mullen et al., 2009). Estimates of the N requirement per unit of lint produced range from 10 to 29 kg N 100 kg^{-1} lint across global cotton production regions (Miley and Oosterhius, 1989; Gerik et al., 1998). Typical application rates in cotton production regions of the United States (U.S.) have reported ranges from 71-220 kg N ha^{-1} (Taylor, 1995). Interestingly, cotton cultivar improvement since 1970 has resulted in cultivars with average lint yield

increases of about 40%, and N uptake and internal nutrient use efficiency increases of approximately 20% (Rochester and Constable, 2015). This suggests that selective breeding for increased yields has indirectly resulted in increased N uptake and iNUE, and could possibly mean that N requirements could be less than previously reported. However, a major factor adding to the variability in estimates of N requirements is the overall susceptibility of N loss through NO_3^- leaching and denitrification, both of which are heavily influenced by unpredictable seasonal environmental conditions and soil texture.

Using in-season N applications that split the total N applied into percentages applied at varying developmental stages is a management strategy that attempts to increase N efficiency by synchronizing soil N concentrations to plant N demand. Cotton nitrogen accumulation is non-linear with the threshold of increase reported to begin at first square, reaching peak accumulation rates at approximately early-mid flower - the developmental stage that accounts for up to 23-45% of the total N accumulation (Armstrong and Albert, 1931; Olson and Bledsoe, 1942; Bassett et al.,1970; Halevy, 1976; Oosterhuis et al., 1983; Halevy et al., 1987; Mullins and Burmester, 1991). Furthermore, N accumulation in the seeds and lint account for up to 43-60% of the total plant N, creating a large N demand sink relatively late in the growing season (Mullins and Burmester, 2010). Geng et al. (2015) reported a yield increase of 106 kg lint ha^{-1} in one of two growing seasons when comparing a split N application (180 kg N ha^{-1} total application rate) of 40% at pre-plant and 60% at first flower compared to 100% of the N applied at pre-plant. Additionally, Yang et al. (2011) observed that when splitting N application rates of 225 kg N ha^{-1} between pre-plant and peak bloom (with a 40%

application rate utilized at first bloom across all treatments), lint yields were greatest when applied at 0-40-60 % split application. Other reports have observed declines in maximal obtainable lint yields when comparing split N applications of 50% at pre-plant and first square, to yields obtained by applying 100% pre-plant N, when utilizing N rates up to 180 kg N ha^{-1} (Reiter et al., 2008). However, splitting N generally resulted in a greater amount of lint produced per kilogram of N applied translating into a greater economic return.

Soil texture has also been demonstrated to influence both the total in-season N rates and the timing of application. Some studies utilize the concept of Economically Optimal Nitrogen Rate (EONR), defined as the N rate that maximizes the value of cotton produced at a given cotton and fertilizer price. The EONR concept is useful for evaluating the impact of N rates across different soil textures which have varying levels of productivity. For example, Scharf et al. (2012) reported a lower EONR for silt and sandy loams when compared to a clay soil. The EONR value was also useful for elucidating the impact of soil type on in-season N applications applied at early square growth stage. This study also evaluated different timings on in-season N applications. No yield differences were observed when N was evenly split (56 kg N ha^{-1}) between pre-plant + early square or pre-plant + early flower in sand and silty loams. However, optimal N split for a clay soil was 1/3 pre-plant, 1/3 early square, and 1/3 early flower (Scharf et al., 2012).

Individually, both N and irrigation management have been examined extensively in cotton; however, less research has examined these management inputs simultaneously as they occur in production systems. The Florida cotton production

environment is characterized by soils high in sand content having excessive permeability which can result in rapid soil water depletion, and the need for supplemental irrigation to mitigate crop production losses due to water deficit stress. High intensity precipitation events can also occur during the growing season resulting in nutrient leaching and the need for in-season nutrient applications to avoid crop nutrient deficiencies. To address these issues there is a need to assess both irrigation and in-season N management to develop management strategies for growers to optimize cotton production by reducing management inputs which may not result in yield improvements.

Dissertation Overview

Irrigation and N management are the two most costly management inputs in U.S. peanut and cotton production systems for avoiding crop production reductions due to abiotic stress. The use of both vegetative RDI and in-season N applications has been documented in the literature to increase the efficiency of irrigation water and N inputs per unit of yield gained. However, the specific amounts of water or N inputs while utilizing these management reduction strategies are dependent on the production environment of interest. To date, no examination of the simultaneous management of both vegetative RDI and N management has occurred in the southeast cotton production system. Although the use of vegetative RDI has been explored in arid peanut production regions of the U.S. (Rowland et al., 2012), it has not been examined in the sandy soils of the humid Southeast. Furthermore, the examination of whole-plant peanut trait responses of disparate peanut genotypes to these irrigation practices has yet to be explored in this environment. The assessment of these genotypic traits responses is critical for identifying germplasm which may be better adapted for a

particular soil hydrologic scenario and utilized in a breeding program. Therefore, the proceeding chapters of this dissertation will address these research needs of determining both genotypic adaptability and efficient water/N management strategies for optimizing resiliency in southeastern U.S. row crop production systems.

CHAPTER 2
EVALUATION OF PEANUT GENOTYPIC POD YIELD ADAPTABILITY TO IRRIGATION MANAGEMENT ACROSS U.S. PRODUCTION REGIONS

Introduction

Over the past 3,500 years, cultivated peanut has expanded into the largest gene pool of the *Arachis* genus including subspecies *fastigiata* and *hypogaea*, which are further separated into the botanical varieties of *vulgaris, fastigiata, peruviana, aequatoriana, hypogaea*, and *hirsuta* (Pasupuleti and Nigam, 2013). Prior to the inception of germplasm collections in 1959, only a narrow portion of this genetic diversity had been incorporated into breeding programs within the U.S. This considerably narrowed the genetic diversity within cultivars such that Knauft and Gorbet (1989) estimated that the ancestral lines of 'Dixie Giant' and 'Small White Spanish-I' were germplasm sources for 100 and 90%, respectively, of all pedigrees in the U.S. Further estimation revealed that both 'Dixie Giant' and 'Small White Spanish-I' contributed 50% of the germplasm to runner market type cultivars prior to 1969 (Knauft and Gorbet, 1989). After the development of 'FloRunner' in 1969, the coefficient of parentage, defined as the probability a random allele at any locus in one cultivar is identical to a descended cultivar (Kempthorne, 1969), was 0.20 and 0.21 for Runner and Virginia market types, respectively. This indicates progress was made in U.S. peanut breeding programs in introducing genetic variability in cultivars developed after 1969.

Development of core and mini-core collections have further advanced the ability of peanut breeding programs to utilize a reduced number of phenotypically diverse genotypes for practical trait screening while still incorporating improved levels of genotypic diversity (Brown, 1989; Holbrook et al., 1993; Upadhyaya et al., 2003;

Upadhyaya et al., 2005; Upadhyaya et al., 2006). The peanut mini-core collection includes 184 accessions representing 10.8% of accessions in the core collection (Upadhyaya et al., 2002), a collection representing the theoretical genetic variation of economically important traits in the U.S. germplasm collection. This has been estimated by using the Shannon-Weaver index, a measure of allelic richness and evenness in a population, which was similar when comparing 13 morphological and agronomic descriptors between the core and mini-core collections (Upadhyaya et al., 2002). Furthermore, the mini-core has been reported to have a wide variation of genotypic pod yield ranging from 93 to 2,287 kg ha^{-1} (Upadhyaya et al., 2002). Although this demonstrates that genetic factors significantly contribute to yield variability among peanut genotypes, it is still imperative to examine other factors that influence this complex, quantitative trait.

Although genotypic variation may have a strong influence on both crop yield and yield components, these traits are of a polygenic nature and are highly influenced by the environment (Cattivelli et al., 2008). In peanut specifically, an analysis of 504 accessions across six environments in Asia resulted in broad sense heritability values of 35.5 and 56.9% for pod yield $plant^{-1}$ and pod yield $plot^{-1}$, respectively (Upadhyaya et al., 2005). This low to moderate range of broad sense heritability highlights the importance of examining phenotypic traits of different peanut genotypes across multiple environments for separating genotype and genotype by environment (GxE) influences to allow for the possible development of cultivars either for specific or broad ranging environments (Mothilal 2012). A further refinement of this concept has occurred through improved understanding that the combination of both management and genetic gains

has contributed to historical increases in yield production: a concept known as genotype by management interaction (GxM) (Tollenaar and Lee, 2002). Therefore, it is critically important to take into account the influence of GxE and GxM across production regions for a given crop as a way to optimize the potential for success in improving crop yield performance.

Considering the environmental variability among the geographical regions in U.S. peanut production accounts for the GxE impact on yield potential. Similarly, accounting for GxM must consider important crop management strategies that may differ among regions. It is likely that GxM contributes to GxE and could be another measurable and manageable component of GxE. One critically important management component that is likely to have strong influence on peanut genotypic yield potential is irrigation. Irrigated peanut acreage has recently accounted for 46 and 95% of the harvested peanut land area in the major peanut producing regions of the southeast and southwest, respectively (USDA-NASS, 2014). Heavy reliance on irrigation across regions regardless of annual precipitation, displays the importance of supplemental water to combat the uncertainty in precipitation patterns that may decrease yield and impact grower profitability. Research in the southeastern U.S. has shown that overall yield increases are observed with irrigation; however obtaining a positive net return on irrigation cost can largely be dependent on peanut prices and crop rotation practices (Lamb et al., 1997; Lamb et al., 2007). It is important to examine the variability of peanut germplasm in response to irrigation to determine the potential for water application to influence yield potential in different genotypes.

The importance of both GxE and GxM to yield potential indicates that the optimal approach in breeding for a crop that is produced across large geographical ranges is a consideration of the genotype by environment by management interactions (GxExM). This approach is particularly important for peanut breeding programs in the U.S. because of the large geographical range of peanut production. Specifically, U.S. peanut production occurs primarily in the states of Florida, Georgia, Alabama, and Mississippi in the southeast; Virginia, North Carolina, and South Carolina in the east; and Texas, New Mexico, and Oklahoma in the southwest. These three regions (southeast, east, and southwest) accounted for an average of 73, 17, and 10% of the total area planted to peanut from 2014-2015, respectively (USDA-NASS, 2015). These regions also encompass extreme variation in disease pressure, temperature range, humidity conditions, rainfall patterns, and growing season duration. For peanut, two wide regional environmental characterizations can be made: 1) the southwestern production areas are characterized by a semi-arid climate with low annual rainfall and high vapor pressure (VPD) deficits; and 2) the southeast and eastern production areas are characterized by humid conditions with high total annual precipitation and, in some years, frequent precipitation events. Although the humid southeastern and eastern regions typically receive more rainfall than the southwestern region, growing season rainfall patterns can be unpredictable and geographically isolated, which when combined with highly permeable soils in some areas, creates a potential need for supplemental irrigation. The susceptibility of yield reductions across both regions due to large ranges of GxExM provides a need to explore the responses of genetically diverse peanut genotypes in an effort to increase yield potential across both environments.

Assessment of GxExM of diverse peanut genotypes produced across regions can be accomplished through the quantification of pod yield stability. Finlay and Wilkinson, (1963) developed an analysis which assesses yield stability across differing environments by plotting individual genotypic yield means against the population mean for specific site years, with the population mean across site locations and years being reflective of the relative yield potential. Thus, the regression coefficient for a specific genotype reflects its stability across varying environmental conditions, with a value of one representing average stability, and regression coefficients below or above one representing stable or unstable yield, respectively. Several studies have used this approach for assessing peanut pod yield stability across environments, as well as genotypic maturity and disease tolerance (Anderson et al., 1989; Adomou et al., 1997; Isleib et al., 2013; Narh et al., 2014). However, to our knowledge this analysis has not been used for examining genotypic peanut pod yield stability across varying environmental regions of the U.S.

Therefore, to evaluate diverse peanut genotypes for their relative GxExM effects on yield (with an emphasis on irrigation management as the most critical M component), we implemented research at multiple locations in the southeast and southwest U.S. utilizing genotypes from the peanut mini-core collection. The overall objective of this research was to evaluate yield stability of these peanut genotypes across sites varying in regional environmental characteristics and irrigation management. We hypothesized that a range of yield stability values would be present in different sites, thus providing important information for breeding efforts aimed at developing regionally-adapted cultivars.

Materials and Methods

Site Information: Field studies were conducted during the years of 2013 and 2014 at: 1) the University of Florida's Plant Science Research and Education Unit near Citra, Florida (Citra) (29° 24' 38" N, 82° 10' 12" W); 2) the USDA-ARS Cropping Systems Research Laboratory near Lubbock, Texas (Lubbock) (33° 41' 38" N, 101° 49' 13" W); and 3) a grower's field near Brownfield, Texas (Brownfield) (33° 01' 55.76" N, 102° 17' 48.52" W). Variation in soil texture occurred across sites: the Florida location was classified as Gainesville loamy sand (Hyperthermic, coated Typic Quartzipsamments); Brownfield was classified as Amarillo-Clovis loamy fine sand complex (Amarillo: Fine-loamy, mixed, superactive, thermic Aridic Paleustalf; Clovis: Fine-loamy, mixed, superactive, mesic Ustic Calciargids); and the Lubbock location was classified as Amarillo-Acuff loamy fine sand (Amarillo: Fine-loamy, mixed, superactive, thermic Aridic Paleustolls; Acuff: Fine-loamy, mixed, superactive, thermic Aridic Paleustolls).

Field Experiments: The experimental design at all locations consisted of a split plot arrangement in a randomized complete block design. The whole plots consisted of irrigation treatments which were applied using a center pivot. The Brownfield and Lubbock locations had four replications, and the Citra location had three replications. For the Brownfield site, adequate irrigation (100%) was based on farmer well capacity, and the low irrigation (50%) treatment was implemented by decreasing the irrigation application frequency. At the Lubbock site, irrigation was set up to replicate the irrigation amounts applied at the Brownfield location, although the low irrigation was applied at the same frequency of the 100% treatment, but involved reducing the irrigation application amount by 50%. Due to the greater probability of precipitation at the Citra

location, and soils with lower water holding capacity, irrigation treatments consisted of plots which received irrigation ranging from 12.7-25.4 mm per application (100%), while the reduced water application treatment was dependent on rainfall alone (rainfed control, RF). Irrigation was scheduled when soil moisture tension at 15 cm soil depth reached -20 to -25 kPa.

Nine peanut genotypes (C76 16, Chico, ICGS 76, COC 041, TMV 2, Serenut 5R, Serenut 6T, ICGV 86015, and ICGV 86388) were planted to the sub-plots across all locations. Five additional genotypes, New Mexico Valencia C (NMVC), FlavorRunner 458, FloRun^{TM} '107', and TUFRunner^{TM} '511', were included at the Citra location. At Citra, four row plots were planted to a length of approximately 4.5 m with 0.9 m row spacing at approximately 1.8 seeds m^{-1} using a two-row planter (Monosem, Inc., Edwardsville, KS). In-furrow applications of Rhizobium inoculum at 1.31 l ha^{-1} (Advanced Biological Marketing, Van Wert, OH) were applied to enhance nodulation. To prevent thrips foliage feeding and the risk of developing *Tomato spotted wilt virus*, phorate at 1.52 kg a.i, ha^{-1} (Amvac Chemical Corporation, Los Angelas, CA) was also applied; azoxystrobin at 0.21 kg a.i. ha^{-1} (Sygenta International AG, Basel, Switzerland) was applied at planting to prevent early fungal infection. For the Brownfield and Lubbock trials, peanut genotypes were planted to the sub-plots in four row plots to a length of approximately 6.0 m and 1.0 m row spacing at approximately 1.8 seeds m^{-1}. In-furrow applications of Rhizobium inoculum at 0.94 l ha^{-1} were made (Optimized Lift, EMD Crop Bioscience, Milwaukee, WI).

At harvest, the lengths of the two center rows of each plot at the Citra location (quantified in length just after peanut emergence) were dug/inverted using a two row

peanut digger (Kelly Manufacturing Co., Tifton, GA). Inverted peanuts were allowed to air dry in the field until the canopy tissue dehydrated; plants were then threshed to remove pods from pegs and foliage using a peanut thresher (Kincaid, Haven Kansas). Pods were collected in burlap bags and dried to approximately 10.5% using a forced air drying wagon. At the Brownfield and Lubbock locations, rows were dug with a two row onion blade and inverted by hand. For the Brownfield trial, inverted plants were allowed to dehydrate in the field for one week. At the Lubbock location, inverted rows were threshed after drying in the field for approximately 6 hours using a peanut thresher (K.E.W., Kingaroy, QLD. Australia). Pods were collected in burlap bags and air-dried to approximately 10.5%.

Evaluation of the impact of water applied: Daily meteorological data were recorded using an automated weather station (Campbell Scientific, Logan, UT; Spectrum Technologies, Inc., Aurora, IL) located within 1500 m of the experimental plots at each site location. Daily weather data collected were used in determining the total water received (rainfall + irrigation) as a percentage of the ET_o (TWRPET). Reference ET_o was calculated using the Penman-Monteith equation (FAO-56 Method) (Monteith, 1965; Allen et al., 1998). To determine how TWRPET might affect critical periods of phenological development due to inherent variability in different sensitivities to water deficit stress in peanut (Reddy et al., 2003), TWRPET was calculated at 0-45, 45-90 DAP, and > 90 DAP. The 45-90 DAP encompasses a development period where pod yield declines are most sensitive to water stress, while >90 DAP is relatively insensitive (Patel and Golakiya, 1988). This approach aids in determining the relative

potential impact of TWRPET on peanut yields; if TWRPET is low during developmental periods when stress sensitivity is high, the impact on yield is predicted to be great.

Statistical Analysis: Statistical analysis was performed using SAS v. 9.4 statistical software (SAS Institute, 2013). PROC GLIMMIX was used to compute analysis of variance (ANOVA) for pod yield traits. The data from each site location was analyzed separately. Random effects of rep nested within year, and rep crossed with irrigation nested within year were included in the model. Normality and homogeneity were visually assessed by graphing the residual distribution, scatter plot of residuals, and Q-Q plot of residuals. Data were pooled over factors if F-test results had a $P > 0.10$. Multiple comparisons were performed using Fisher's Protected Least Significant Difference (LSD) at a $P < 0.10$ probability level.

Regression analysis was performed using PROC REG (SAS v. 9.4; SAS Institute, 2013). The regression analysis at the Citra location was performed by predicting the response of genotypic pod yield to increasing total water received in the two site years and irrigation treatments as indicated by the regression coefficient (b). The amount of total water received for the two irrigation treatments across the two study years was 574, 742, 812, and 951 mm so yield responses per unit of water are within the total water received range of 574-951 mm. Genotypic yield stability was performed for each genotype by plotting the average genotypic yield for each site year and irrigation treatment against the population mean for each site year and irrigation treatment, as according to Finlay and Wilkinson, (1963).

Results

Brownfield, TX

Total precipitation received was 33 and 345 mm in 2013 and 2014, respectively (Figure 2.1), requiring application of more irrigation water in 2013 than 2014 due to the substantially drier conditions. In 2013 were 260 and 340 mm was applied in the 50 and 100% irrigation treatments, respectively; while in 2014, 122 and 231 mm was applied in the 50 and 100% treatments, respectively. Total water received as a percentage of ET_o (TWRPET) was 31 and 39% in the 50% and 100% irrigation treatments in 2013, respectively. In 2014, the TWRPET was 54 and 66% in the 50 and 100% irrigation treatments, respectively.

The effect of low yearly TWRPET may have been even more detrimental in 2014 than 2013 due to the pattern of low levels of TWRPET during critical developmental stages in 2014. When examining TWRPET at the critical period of phenological development between 45-90 DAP when pod development is occurring, the TWRPET in 2013 was 41 and 62% in the 50 and 100% irrigation treatments, respectively; while in 2014 for this same time period, was 21 and 33% in the 50 and 100% irrigation treatments, respectively. These results indicate that the peanut genotypes experienced a greater degree of water stress during the critical 45-90 DAP development stage in 2014 versus 2013. The interaction of site year with irrigation on peanut yields also supports the argument that the level of water stress during the 45-90 DAP period in 2014 contributed to a large yield decline (Table 2.1). Overall, average pod yields were reduced by 561 kg ha^{-1} comparing the 2014 growing season to 2013 (Table 2.2). Although the 100% irrigation treatment yielded similarly in both years, the 50% irrigation treatment was 1351 kg ha^{-1} lower in 2014 than 2013. The TWRPET results for the 50%

treatment suggest that the likely cause of this yield loss was the water availability during the 45-90 DAP period in 2014.

Lubbock, TX

Precipitation received in 2013 and 2014 was 229 and 399 mm, respectively (Figure 2.2). Irrigation amounts in 2013 were 108 and 191 mm in the 50 and 100% irrigation treatments, respectively; while in 2014, irrigation amounts were 159 and 305 mm in the 50 and 100% treatments, respectively. The yearly TWRPET when averaged across irrigation treatments in 2013 and 2014 was 36 and 67%, respectively. The distribution of rainfall was also different among the years, with a lower total received in 2013, but a more even distribution than in 2014. In 2014, approximately half of the rainfall was received after 90 DAP, resulting in a TWRPET from 90 DAP until harvest of 127%, as compared to 20% for that same time period in 2013.

Late season water deficit in 2013 likely contributed to lower yield levels as compared to 2014. However, there was year by genotype interaction for yield, indicating that genotypes differed in their relative sensitivities to differing water conditions between the dry year of 2013 and the wet year of 2014 (Table 2.1). Only one genotype, ICGV 76, had greater pod yield in 2013 than 2014; while yield of others was less in 2013 compared to 2014, including Chico, ICGV 86388, and Serenut 5R. All other genotypes yielded similarly in both years (Table 2.2).

Citra, FL

Total precipitation amounts in 2013 and 2014 were 729 and 544 mm, respectively, at the Citra location (Figure 2.3). Irrigation amounts in 2013 were 12.7 and 222 mm in the 50 and 100% irrigation treatments, respectively; while in 2014, irrigation amounts were 31 and 268 mm in the 50 and 100% treatments, respectively. An

irrigation amount applied in the rainfed treatment for both years was to ensure optimal germination and crop establishment. The high amounts of precipitation in 2013 resulted in a seasonal average TWRPET of 134 and 172% in the rainfed and the 100% irrigation treatment, respectively. The TWRPET was relatively lower in 2014 with an amount of 90% in the RF treatment and 127% in the 100% treatment.

In 2013, pod yield did not increase with irrigation, likely due to adequate precipitation amounts (729 mm) (Table 2.3). Pod yields increased by 588 kg ha^{-1} in 2014 when comparing the 100% irrigated treatment to the rainfed. When averaged across both years, genotypes differed in their yield response to irrigation, with increased pod yields in the 100% treatment for Georgia-06G and NMVC; and decreased pod yield for genotype ICGS 76. Genotypes Flavorunner 458, FloRun^{TM} '107', and C76 16 had a trend of decreased pod yields for each additional increase mm of total water received. The regression coefficient predicting genotypic pod yield response per unit of total water received over the two site years was negative for Flavorunner 458, FloRun^{TM} '107', and C76 16; while those with a positive relationship were NMVC, Chico, and COC 041 (Table 2.4).

Genotypic Yield Stability

Yield stability was assessed across all environments in an effort to determine which genotypes may have characteristics allowing them to maintain yield across environments. Stability was measured by examining how the regression coefficient (b) varies from one, which would be an indicator of average stability. Stable ($b \leq 0.90$) genotypes are ones having broad range adaptability; while unstable ($b \geq 1.10$) genotypes have specific adaptability (Finlay and Wilkinson, 1963). Regression coefficients when modeled for each genotype across all irrigation treatments and site

years ranged from 0.87 to 1.3 (Table 2.5). Genotypes C76 16, Chico, and ICGS 766 had b < 0.9; while genotypes ICGV 86015, ICGV 86388, and TMV 2 had b ranging from 0.9 to 1.0. The genotypes Serenut-5R, Serenut-6T, and COC-041 had b > 1.1.

Discussion

A strong main effect of genotype (G) and its interaction with year (GxY) occurred within all locations indicating that genetics and yearly environmental variation are predominant factors influencing yield potential. This has been demonstrated for peanut by Upadyaya et al. (2005), who reported a broad sense heritability of 57% in a mini-core collection conducted across environments in Asia. In the current study, few differential genotypic pod yield responses to irrigation occurred at each site location indicating a relatively low GxM interaction on pod yield. However, this result may have also been partly due to the lack of dry conditions and substantial differentials in the amount of total water received across most site years.

There were instances in this study where water differentials occurred in total water received among irrigation treatments. For example, at the Brownfield location, the average (across both years) differential in TWRPET between the 50% and 100% irrigation treatment during the 45-90 DAP period, when peanut pod yields are most susceptible to water deficit, was 31 and 48%, respectively. This likely contributed to the 52% decrease in pod yield compared to the 100% treatment. At this site, there was also no irrigation by genotype (GxM) interaction. This lack of a GxM or GxMxY interaction at the Brownfield location demonstrates that, at more severe levels of water stress, all genotypes were susceptible to yield reductions, suggesting that the variability of peanut genotypic tolerance to water stress is more likely to occur at moderate stress levels. Upadhyaya et al. (2002) reported a Shannon-Weaver index for pod yield of 0.60 and

0.17 during rainy and post-rainy season, respectively. Although a pod yield reduction occurred in the 50% irrigation treatment in both seasons at Brownfield in the current study, the yield decline was greater in the 2014 growing season, despite a greater average TWRPET over the growing season. This was likely due to the severity of water stress received during the 45-90 DAP period, a developmental period in which water stress most greatly impacts pod yields (Rao et al., 1985; Patel and Golakiya, 1990). The Lubbock site in the dry year of 2013, also experienced a more severe water differential, but in this case, the genotype ICGS 76 increased pod yield production. This same genotype at the Citra site also had increased pod yield in the rainfed compared to the 100% treatment. This response indicates that ICGS 76 may have drought tolerance under more severe water stress. Other studies have demonstrated ICGS 76 to exhibit both heat tolerance and increased osmotic adjustment capacity under drought which may contribute to its ability to withstand severe water scarcity (Selvaraj et al., 2011; Padmavathi and Rao, 2013).

Differential genotypic yield responses to irrigation (GxM) was present in the current study under mild levels of water deficits at the Citra location. The relationship between genotypic pod yield and total water received showed both positive and negative responses among cultivars. Genotypes which had a positive relationship between pod yield and total water received were NMVC, Chico, and COC 041, indicating a particular sensitivity in pod yield production to fluctuations in total water received. There is evidence of this relationship at least for COC 041 when examining its stomatal sensitivity to soil drying. Kotapalli et al. (2009) observed greater and faster reduction of stomatal conductance to the onset of soil drying in COC 041 compared to

COC 166. However, COC 041 also exhibited rapid stomatal opening (leading to increased photosynthesis) following soil re-wetting, indicating an overall greater sensitivity for this genotype to soil water conditions. Genotypes with more sensitive stomatal control are likely to have positive pod yield responses in environments with intermittent drying and wetting cycles.

When total water received exceeded 544 mm at the Citra location, the genotypes FloRun^{TM} '107', C76 16, and Flavorunner 458 responded negatively to each additional mm of water received, indicating a possible maladaptation to sites subject to periods of excessive precipitation. Yield declines could be due to genotypic physiological responses to excessive soil water content in combination with increased disease susceptibility. Reports have indicated the susceptibility of Flavorunner 458 to *Sclerotinia minor* (Woodward et al., 2015). Interestingly, genotypes in the current study that had positive pod yield responses to total water received were of *fastigiata* descent, and negative pod yield responses were of *hypogaea* descent. These results suggest differences in inherent adaptive mechanisms between the two subspecies which may be related to their origin. Krapovickas and Gregory (2007), reported that evidence supports that subspecies *hypogaea* had its origin in southeast Bolivia, whereas subspecies *fastigiata* differentiated itself further north, possibly Peru. Finally, the sensitivity of peanut genotypes to more than adequate levels of soil water content is novel, as most studies have been focused on water scarcity conditions only. These results demonstrate the importance of examining genotypic tolerance to both drought and excessive soil water content for improving cultivar development across peanut producing regions.

Yield stability assessed by examining the interaction of GxExM varied considerably among peanut genotypes, with Serenut 6T, Serenut 5R, and COC 041 demonstrating the lowest stability across all environments. In production environments where the population pod yield mean was low, these genotypes tended to have the lowest yields, while large relative yield increases were observed in the production environments with greater population pod yield means. These genotypes are all of subspecies *fastigiata* descent, further validating their specific adaptability to greater potential yield production scenarios. However, their yields in high yielding environments were still lower than the population mean. The genotypes C76-16, Chico, and ICGS 76 performed the best across all production environments by possessing both high yield stability and overall yield levels. These genotypes have also been documented to have physiological responses which deem them as being water-deficit stress tolerant (Ketring et al., 1990; Ratnakumear et al., 2009; Dang et al., 2013). Therefore, this germplasm may be best suited for production regions that are characterized as either arid or humid environments.

Conclusion

The stability analysis of this study showed variation in the genotypic pod yield responses to irrigation and environment (GxExM). Genotypes C76 16, Chico, and ICGS 76 in this study had the greatest pod yields and stability indicating their broad adaptability. In contrast, COC 041, Serenut 5R, and Serenut 6T had relatively low yield stability across site locations and irrigation management, demonstrating their specific adaptability to the site locations with the greatest population pod yield means. The interaction of genotype by irrigation management (GxM) varied among the site locations and years which was likely due to the range in severity of water stress conditions

among sites. At the Brownfield location, severe water stress resulted in a pod yield reduction across all genotypes in the 50% irrigation treatment. This result, with a lack of genotype by irrigation interaction (GxM) implies that genotypic pod yield variation is most likely to occur at more mild levels of water stress. This is further supported by the genotype by irrigation interaction (GxM) which occurred at the Citra location, where greater amounts of TWRPET occurred. At this location, both C76-16 and ICGS-76 responded negatively to increasing total water received, indicating some susceptibility of decreasing pod yields in environments which receive high amounts of precipitation. In contrary, genotypes of NMVC, Chico, and COC 041 had a positive pod yield response to increasing total water received, demonstrating greater specific adaptability to the humid Citra environment. Further research which examines whole plant trait responses relating to genotypic pod yield may provide greater understanding and strengthen the assessment of both broad and specific adaptability of divergent peanut genotypes to environmental conditions and irrigation management.

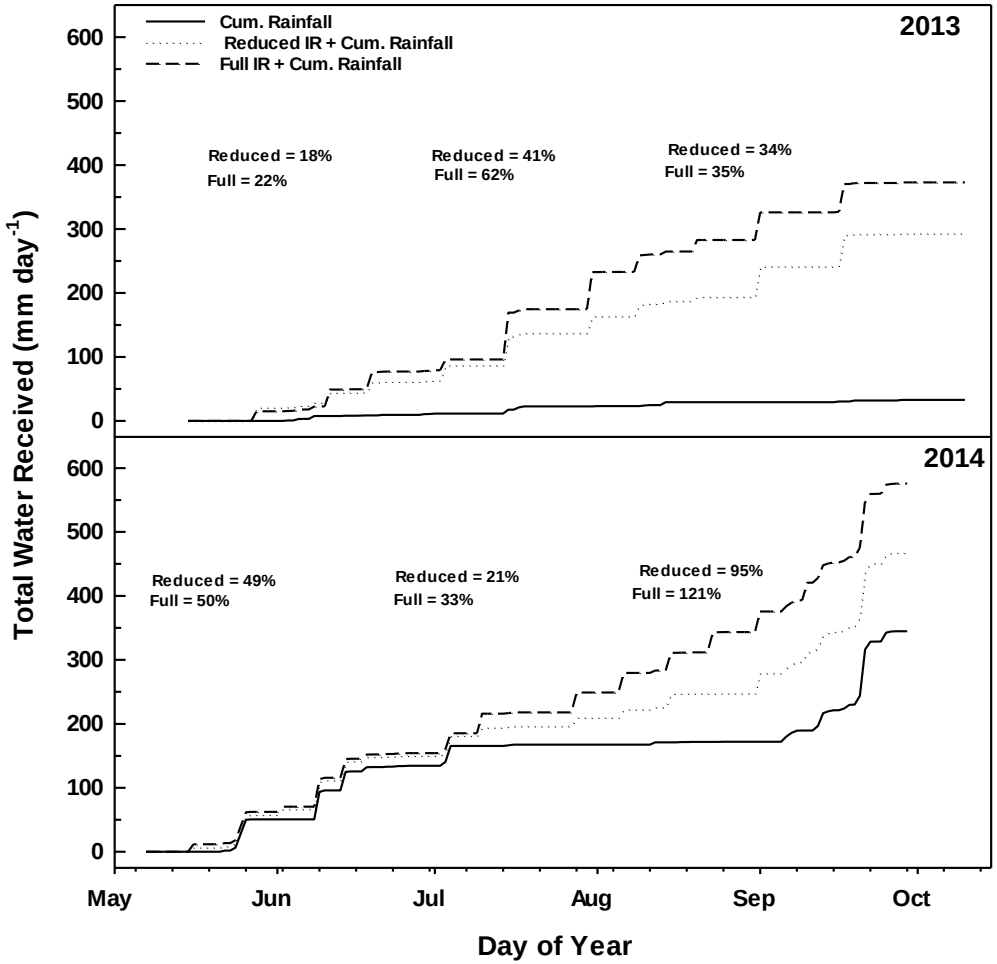

Figure 2-1. Precipitation and total water received for each irrigation treatment at the Brownfield, TX location for 2013 and 2014.Shaded colors represent developmental time of 0-45, 45-90, and >90 days after planting. Percentages during developmental time periods for 100% and 50% irrigation treatments are the total water received as a percentage of ET_o.

Table 2-1. Analysis of variance (ANOVA) for pod yield response variables at each site location in 2013 and 2014.

Pod Yield ANOVA by Location			
Effect	Brownfield	Lubbock	Citra
YEAR	0.0159	0.5719	0.0336
IR	<0.0001	0.1265	0.3556
YEAR*IR	0.0034	0.1761	0.0657
GENO	<0.0001	<0.0001	<0.0001
YEAR*GENO	<0.0001	0.0043	<0.0001
IR*GENO	0.3543	0.4576	0.0613
YEAR*IR*GENO	0.3393	0.8480	0.6462

†Abbreviations: Year, growing seasons of 2013 and 2014; IR, irrigation treatment; GENO, genotype treatment.

Table 2-2. Average genotypic pod yield for the Brownfield, Lubbock, and Citra location in 2013 and 2014.

	Location		
Genotype	Brownfield	Lubbock	Citra
2013	---------- kg ha^{-1} ----------		
C76 16	4811a	3508a	2921bc
Chico	4113b	2319bc	3261ac
ICGS 76	3663bc	3387a	3170ab
COC 041	3331cd	1534de	3296bc
TMV 2	3118ce	1741cd	3938d
Serenut 5R	3147ce	1055e	2703ab
Serenut 6T	2928de	1572ce	3732bc
ICGV 86015	2735e	2070b	3103a
ICGV 86388	1672f	1203ce	2119cd
Mean	3280	2043	3138
2014			
C76 16	2850ac	3006a	6297a
Chico	3158a	2242b	2641de
ICGS 76	3232a	1990bc	4085b
COC 041	2771bc	1080e	2238e
TMV 2	2336bc	1329de	2402de
Serenut 5R	1827c	1255de	3852d
Serenut 6T	2569ab	1406de	3803bc
ICGV 86015	2529ab	1809bd	3178b
ICGV 86388	2393ac	1462cd	3138cd
Mean	2629	1731	3515

†Different letters within each column represent significance using Fischer's Protected Least Significant at $P < 0.10$.

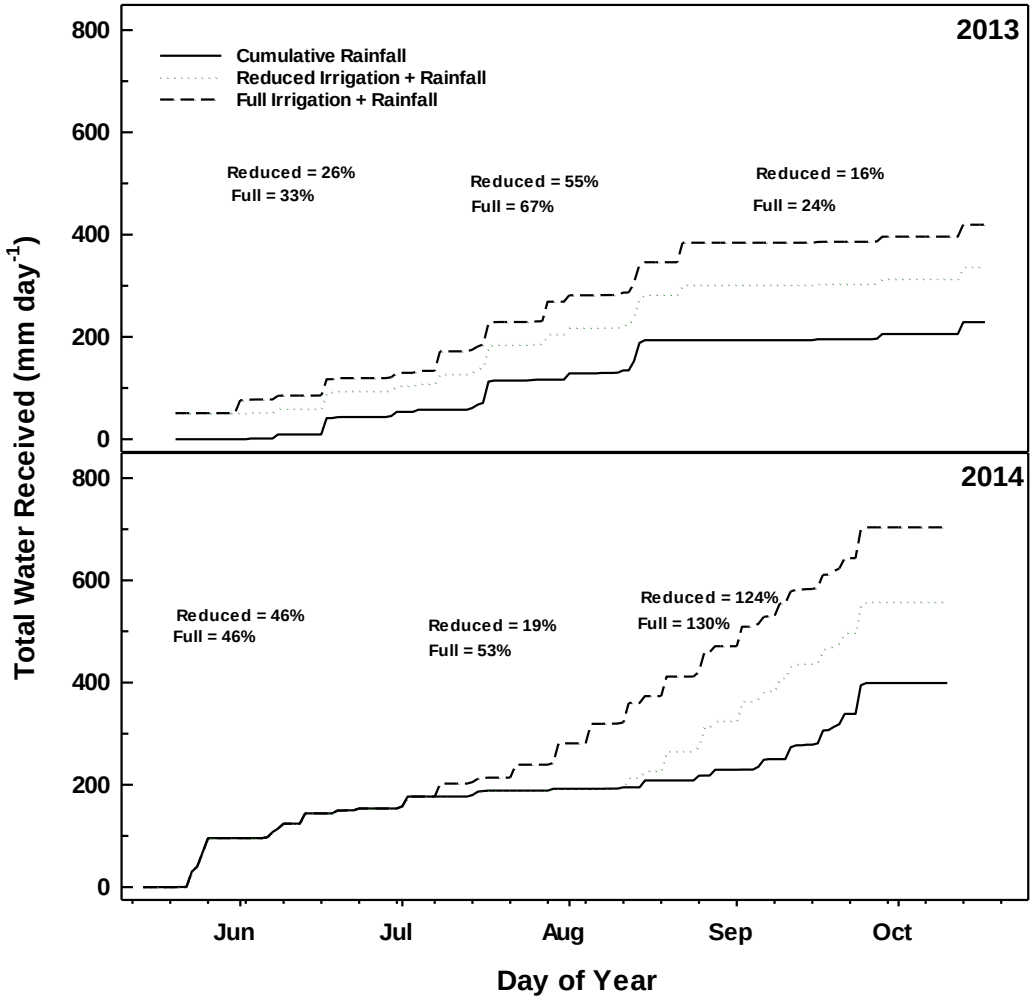

Figure 2-2. Precipitation and total water received for each irrigation treatment at the Lubbock, TX location during in 2013 and 2014.Shaded colors represent developmental time of 0-45, 45-90, and >90 days after planting. Percentages during developmental time periods for 100% and 50% irrigation treatments are the total water received as a percentage of ET_o.

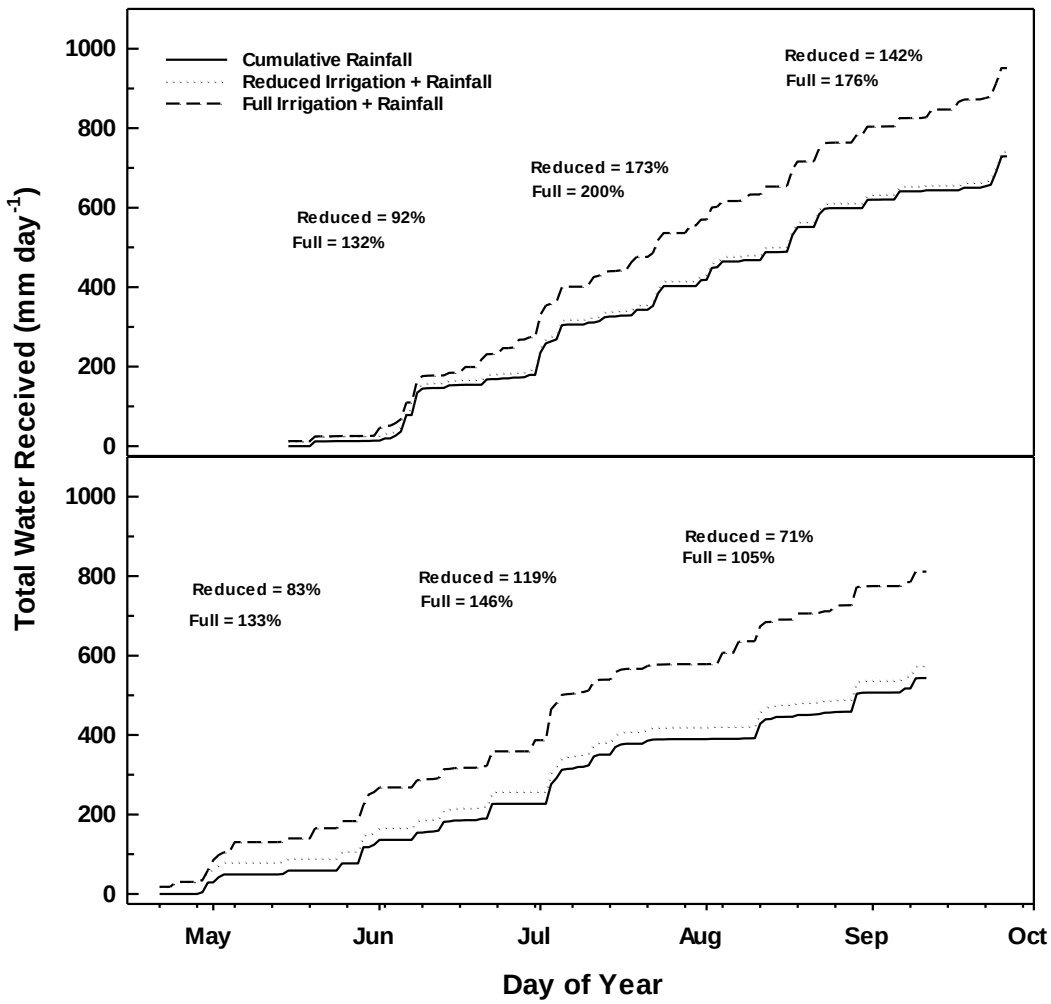

Figure 2-3. Precipitation and total water received for each irrigation treatment at the Citra, FL location during in 2013 and 2014.Shaded colors represent developmental time of 0-45, 45-90, and >90 days after planting. Percentages during developmental time periods for 100% and RF irrigation treatments are the total water received as a percentage of ET_o.

Table 2-3. Average (n=3) genotypic pod yield for the Irrigated (100%) and rainfed treatments at the Citra, FL location.

Citra Location		
Genotype	Irrigated	Rainfed
	------- kg ha^{-1} -------	
Georgia 06G	5905a†	5152b
C76 16	4369a	4968a
Chico	3187a	2597a
COC 041	2920a	2380a
Flavorunner 458	3226a	3968a
FloRun™ '107'	4830a	5472a
ICGS 76	3332b	4228a
ICGV 86015	4141a	3697a
ICGV 86388	3030a	2756a
NMVC	2772a	1857b
Serenut 5R	3565a	3133a
Serenut 6T	3560a	3293a
TMV 2	2343a	2136a
TUFRunner™ '727'	5510a	5049a
Mean	3764	3621

†Different letters within each row represents significance using Fischer's Protected Least Significant at $P < 0.10$.

Table 2-4. Regression analysis of pod yield response for increasing mm of total water received at the Citra, FL location. The regression coefficient (b) represents genotypic pod yield response for total water received across both years and irrigation treatments.

Genotype	Citra Yield Versus Total Water Received Regression		
	Regression Coefficients (b)	P-value	Model R^2
Georgia 06G	- 2.1	0.5773	0.04
FloRun™ '107'	-6.9	0.0293	0.47
TUFRunner™ '727'	-0.96	0.6619	0.03
C76 16	- 8.9	0.0696	0.35
NMVC	5.1	0.0153	0.54
Chico	3.2	0.0605	0.37
ICGS 76	-4.2	0.1156	0.28
COC 041	3.7	0.0049	0.65
TMV 2	0.08	0.9359	0.00
Serenut 5R	2.8	0.1130	0.28
Serenut 6T	-0.62	0.6866	0.02
ICGV 86015	1.2	0.2251	0.18
ICGV 86388	0.01	0.9942	0.00
Flavorunner 458	-6.8	0.0091	0.43

Table 2-5. Average genotypic pod yield and stability across all three site locations. Regression coefficient (b) represents the pod yield stability (GxExM) which is the average pod yield for each genotype versus the average pod yield for each site location, year, and irrigation treatment.

Genotype	Mean	Pod Yield and Stability Regression Coefficients (b)	R^2
C76-16	3814	0.88	0.25
ICGS-76	3289	0.90	0.42
Chico	2880	0.90	0.52
ICGV-86015	2752	0.93	0.49
Serenut-6T	2429	1.12	0.88
Serenut-5R	2268	1.30	0.80
COC-041	2233	1.14	0.71
TMV-2	2143	0.90	0.76
ICGV-86388	2009	0.87	0.59
Mean	2646	0.99	0.60

†All regression coefficients are significant at a $P < 0.01$.

CHAPTER 3
ASSESSMENT OF ABOVE- AND BELOW-GROUND TRAITS INTRINSIC TO DISPARATE PEANUT GENOTYPES FOR DETERMINING ADAPTABILITY TO SOIL HYDROLOGIC CONDITIONS

Introduction

Since 1950, peanut yield has increased approximately 46 kg ha^{-1} yr^{-1} (NASS, 2017). Major contributions to yield advances during this time have been attributed to the advancement of crop protection pesticides, disease tolerance, and the development of cultivars with greater yield potential (Isleib et al., 2001). Increasing yield potential by breeding new cultivars has been attributed to increasing numbers of reproductive structures, reproductive efficiency (percentage of flowers resulting in pods and seeds), and seed weight (Coffelt et al., 1989; Seaton et al., 1992; Haro et al., 2013). An interesting historical change in global production practices occurred when the preference shifted from growing cultivars with erect canopy growth habits of *fastigiata* descent to cultivars with procumbent canopy growth habits of *hypogaea* descent (Haro et al., 2013). This shift to cultivars with procumbent canopy architectures has been linked to increases in LAI and light attenuation within the canopy, contributing to an overall increase in peanut above ground biomass and crop harvest index (Haro et al., 2017). Although there is much documentation on the influence peanut breeding has had on above ground traits, to our knowledge, no evidence exists on how peanut breeding trends have influenced root architecture. Changes in root architecture may also have been driven in commercial production by the shift from cultivars of *fastigiata* descent to ones of *hypogaea* descent. Knowledge of subspecies root system architecture (RSA), and the relationship of this trait to above-ground partitioning could be critical in genotypic selection for improving water deficit stress resiliency.

Root trait responses that improve soil water acquisition and possibly reduce yield loss from water stress have been documented in the peanut literature. Many studies have reached similar conclusions showing that peanut genotypes with inherently deep root architectures, or that respond to water stress by proliferating roots deeper into the soil profile, have relatively greater pod yields, and under water stress conditions, are deemed as more water stress tolerant (Rucker et al., 1995; Songsri et al., 2008; Jongrungklang et al., 2011; Jongrungklang et al., 2012). These results suggest that water stress tolerance traits are attributed to not only a larger root system, but to proliferation of roots deeper into the soil profile. Further, the study conducted by Jongrungklang et al. (2012) observed a negative relationship between pod yield and percent root length density (%RLD) in the top 30 cm of the soil when water stress was imposed during mid-season development. This indicates that there may be a trade-off between greater root production and yield under stress conditions.

The interaction of genotype with environment and/or management has made it difficult to directly utilize the results of physiological studies on water stress in breeding programs (Cattivelli et al., 2008) because the utility of a physiological adaptation depends on the specific environmental or management conditions the crop experiences. Water stress impacts vary in relation to the timing of stress with phenological development, the rate at which soil drying occurs to achieve water stress conditions, and the severity of water stress conditions imposed. Therefore, it is critical that experimental approaches to evaluate germplasm traits relative to water stress be consistent with the intended typical stress severity and timing for the region for which a specific cultivar is developed (Sinclair, 2011). Another critical factor when studying

water stress resilience is to distinguish the difference between biological and agronomic water stress tolerance. For example, although relative genotypic crop survival under severe water stress conditions may demonstrate water stress tolerance in a biological sense, survival of crop plants is irrelevant in a commercial setting because it does not necessarily preserve harvestable yield and therefore economic viability for the farmer (Sinclair, 2011).

Therefore, experiments that focus on trait assessment for improving water stress tolerance must be conducted within the region of interest, and under water stress conditions that are relevant to the particular deficit conditions commonly experienced in the area where the cultivar is to be produced. This ensures the development of cultivars with some level of local and relevant stress adaptation. One particular environment relevant to peanut production is the southeastern U.S. which encompasses an agronomic region known as the “peanut belt” because the majority of the U.S peanut hectarage is planted in this area (NASS, 2015). The climate in this region is characterized as humid, where the annual precipitation exceeds evaporation, and average seasonal precipitation is often greatest during the summer months coinciding with peanut production (Henry, 2005; SERCC, 2017). A large percentage of this region has soils with high sand content and low organic matter, characterizing them as well-drained with little water holding capacity. Because of these edaphic conditions, water scarcity stress can occur quickly in periods of less frequent precipitation due to rapid soil water depletion; but dry periods are often intermittent due to the high probability of summer precipitation. However, in some drought prone years, the duration of water stress can be prolonged and cause more severe water scarcity stress. Mitigating the

risk of water scarcity in this region has led to an irrigation infrastructure on 60% of the harvested peanut hectarage, thus reducing the duration and the severity of crop water scarcity compared to rain fed production systems (NASS, 2014). However, both irrigated and rainfed production scenarios of this region have a heavy reliance on in-season water supply from precipitation.

An irrigated production environment also allows for more control of the level of water scarcity stress experienced by the crop over the course of phenological development. For example, irrigated growers may choose to utilize regulated deficit irrigation (RDI), an irrigation practice designed to reduce the amount of water application below the full water requirement for optimum plant growth (Chai et al., 2016). Developmental stage-based RDI has been successful for peanut, particularly the use of reduced amounts of irrigation during early season vegetative growth, thus eliciting increased root proliferation deeper into the soil profile without reducing pod yields (Rowland et al., 2012; Thangthon et al., 2016). However, root distribution and pod yield responses have been reported to vary across peanut genotypes demonstrating variation in phenotypic plasticity (Jongrungklang et al., 2011). Furthermore, the phenotypic plasticity to increase root distribution deeper into the soil profile during vegetative water stress is likely heavily influenced by environmental conditions. This is of particular importance since success using vegetative RDI has been documented in arid environments (Rowland et al., 2012), or under controlled conditions with rainout shelters (Jongrungklang et al., 2011; Thangthon et al., 2016). In both scenarios, total water received is primarily through irrigation in contrast to peanut production in humid

environments where stochastic precipitation is the primary water source, and water stress is more likely to be mild.

Major whole plant traits governing crop yield production under non-stressed conditions are the amount of incident solar radiation which is transformed into chemical energy and partitioned into harvestable tissue (Hay and Porter, 2006). Leaf area index (LAI) largely influences the amount of intercepted solar energy, and therefore is positively related to dry matter production when optimal growing conditions are present (Hay and Porter, 2006). However, under conditions of water deficit stress, stomatal closure reduces the amount of CO_2 substrate for photosynthesis which often reduces biomass production. A trait which could possibly mitigate water deficit stress is the RSA of a crop that includes deep rooting ability, allowing water acquisition from deep soil strata (Lynch and Wojciechowski, 2015).

By characterizing LAI, RSA, and pod yields of peanut genotypes varying in subspecies descent, knowledge of innate carbon partitioning can be obtained which could be useful for germplasm selection. Further evaluating these traits under various irrigation management strategies provides additional information on phenotypic plasticity which could possibly be utilized in determining genotypic adaptability to water deficit conditions, and possible irrigation management strategies which can be utilized for a particular production region. Therefore, the objectives of this field research trial were to: (i) assess phenotypic variation in RSA, LAI, and their relationship to pod yield among genotypes classified into peanut subspecies *hypogaea* and *fastigiata*; and (ii) characterize the possible phenotypic interactions of these genotypic traits with environmental variability under various irrigation management strategies relevant for a

humid climate. We hypothesized that genotypic differences in root system architecture exist among subspecies of *hypogaea* and *fastigiata* descent which would provide more or less resilience to water stress conditions, and that pod yield production would be negatively related to belowground traits due to trade-offs in carbon allocation between root and shoot systems. Furthermore, we suspected that above- and below-ground genotypic traits would interact with the range of irrigation treatments implemented in this study because of the disparate descents of these genotypes which may be adapted to different soil hydrological conditions. The information gained from this study will provide knowledge of both above- and belowground biomass partitioning between subspecies *hypogaea* and *fastigiata* that could be used for: i) selecting germplasm best adapted to a range in hydrologic conditions; and ii) evaluating the feasibility of utilizing reduced irrigation management strategies in humid climate peanut production regions.

Materials and Methods

Site Characterization: Field studies were conducted during 2015 and 2016 at the University of Florida's Plant Science Research and Education Unit (PSREU) in north-central Florida (29° 24' 38" N, 82° 10' 12" W). The soil is classified as Candler sand (Hyperthermic, uncoated Lamellic Quartzipsamments). In both years, peanut was planted into a peanut-cereal rye-cotton rotation. Soil preparation consisted of conventional tillage approximately five weeks before planting. Immediately prior to planting, the field was surface tilled using a field cultivar with S-tine sweeps. Seeding density of all peanut genotypes was 20 seeds m^{-1} with a row spacing of 0.91 m. Plots consisted of four rows 7.5 m long. Planting was accomplished with a two-row Monsem® vacuum planter (Monosem Inc., Edwardsville, KS). Rhizobium inoculum (1.31 l ha^{-1}; Advanced Biological Marketing, Van Wert, OH), and azoxystrobin fungicide (0.21 kg a.i.

ha^{-1}; Sygenta International AG, Basel, Switzerland) were applied in the seed furrow at planting. A soil test was conducted approximately six weeks prior to planting, and recommended nutrients were broadcast surface applied immediately after planting. Daily meteorological data was recorded using an automated weather station located within 1500 m of the experiment.

Experimental Design: The experimental design consisted of a split plot arrangement in a randomized complete block design with four replications. The whole plots were irrigation treatments consisting of: (i) 1.9 cm per application (100%); (ii) a primed acclimation (PA) treatment consisting of 1.1 cm of water per application until mid-bloom and then 1.9 cm of water for the remainder of the season (60%PA); (iii) 1.1 cm of water per application for the entire season (60%); and (iv) a rainfed control (RF). Irrigation was scheduled when tensiometers (Irrometer, Riverside, CA) placed in the 100% irrigation treatment of each replication reached an approximate average soil water tension of -25 kPa at a soil depth of 30 cm. Soil water tension level for irrigation scheduling was based on an estimated level of 50% maximum allowed depletion of -20 kPa for a sandy soil (NRCS, 2005). Tensiometer placement at 30 cm was selected as half of the estimated effective root zone. The effective root zone was a conservative estimate based on the FAO56 recommendation range of 0.5-1.0 m (Allen et al., 1998). The full irrigation (100%) amount (1.9 cm) was over estimated to ensure replenishment of soil water to a depth of 30 cm using an available water capacity of 0.04 cm H_2O cm $soil^{-1}$ (NRCS, 2017) assuming an irrigation efficiency factor of 0.80.

Irrigation was applied using a lateral system equipped with variable rate irrigation (VRI) technology (Lindsay Corporation, Omaha, NE). Sub-plots consisted of four

genotypes, two Valencia genotypes (*Arachis hypogaea* L. subsp. *fastigiata* Waldron var. *fastigiata* Waldron) COC 041 (PI 493631) and New Mexico Valencia C (NMVC) (Reg. No. 24, PI 565461), and two runner (*Arachis hypogaea* L. subsp. *hypogaea* L var. *hypogaea* L.) commercial cultivars FloRun™ '107' (Reg. No. CV-127, PI 663993) and TUFRunner™ '511'(Reg. No. CV-131, PI 674432).

Crop Measurements: Mini-rhizotron tubes were used to examine RSA development over the growing season. Mini-rhizotron tubes were installed directly under and parallel to the row at a 45° angle to the soil surface immediately after crop emergence using a hydraulic powered coring machine (Giddings Machine Company, Windsor, CO). At each measurement date, images were captured at 13.5 mm increments (resulting typically in 85-109 image frames) along the mini-rhizotron tubes using a BTC 100X video camera and BTC I-CAP image capture software (Bartz Technology Corporation, Carpinteria, CA). Total root length (TRL) and total surface area (TSA) analysis were conducted using WinRHIZO Tron software (Regent Instruments Inc., Quebec, Canada) by hand tracing of root segments within each image frame. Total length on each image frame was computed by estimating the linear length of each root segment. Total surface area was the product of total length and the cross sectional circumference (CSA) (CSA=2Πr) calculated by estimating the root diameter being equal to the measured diameter of the projected root segment. In both years, root image sampling dates occurred at 21, 28, 35, 49, and 75 days after planting (DAP).

Measurements of leaf area index (LAI) were recorded at 28, 35, 49, 64, and 77 days after planting (DAP) in both site years (LI-COR, Lincoln, Nebraska). A single measurement of LAI for a plot was conducted by taking one above and four equidistant

readings below the canopy as a perpendicular transect between the rows with the sensor view parallel to the row and repeating this sequence with the sensor view perpendicular to the row to gain greater spatial averaging. A lens cap cover with a 45° angle was used on all measurement dates. Measurements were taken in early morning with a cast shadow over the plot area to prevent diurnal bias and underestimation of LAI values.

Plot lengths were measured after peanut emergence to determine the harvest length of the two center rows. Harvest was timed to optimal maturity as was determined by removing the exocarp and revealing the mesocarp layer of the pod using a pressure washer (Williams and Drexler, 1981). Pods were classified as either immature or mature by their mesocarp color; pods which were brown/black were classified as mature pods. When approximately 80% of the pods were mature, the peanut plants were dug/inverted using a two row peanut digger (Kelly Manufacturing Co., Tifton, GA). Inverted peanuts were allowed to dry in field until the canopy tissue dehydrated. Following this drying period, peanut pods were separated from the pegs using a hand thresher (Kincaid, Haven, Kansas), collected in burlap bags, and dried to approximately 10.5% using a forced air drying wagon. Each peanut market type had different digging/inversion dates in both years due to differences in maturity class. In 2015, the Runner and Valencia market types were inverted at 131 and 102 DAP, respectively. The 2016 inversion dates for the Runner and Valencia market types were 142 and 111, respectively.

Statistical Analysis: Statistical analysis was performed using SAS v. 9.4 statistical software (SAS Institute, 2013), with the GLIMMIX procedure computing an analysis of variance (ANOVA). Random effects of rep nested within year, and rep

crossed with irrigation nested within year were included in the model; all other factors were considered fixed. Repeated measures ANOVA was performed on measurements that were repeated over the growing season (TRL, TSA, and LAI). Covariance matrix structure was specified for repeated measure ANOVA's by choosing the structure with the lowest Akaike Information Criterion (AIC). A heterogeneous autoregressive (ARH1) covariance matrix structure was used for TSA and TRL measurements. An autoregressive (AR1) covariance matrix structure was used for LAI. Normality and homogeneity were visually assessed by graphing the residual distribution, scatter plot of residuals, and Q-Q plot of residuals. Total root length and TSA data were heteroscedastic so a square root transformation was performed prior to running the repeated measures ANOVA. Data were pooled over years and experimental treatments when appropriate as indicated by a non-significant F-test for all traits measured. Assessment of RSA was performed by summing parameters across 20 cm increments to a total soil depth of 80 cm. Root parameters were summed across the entire 80 cm sampling depth for determining main and interactive effects not including the soil depth factor. Multiple comparisons significance was determined using Fisher's Protected Least Significant Difference (LSD) at the $P < 0.05$ probability level.

To assess the relationship between pod yield and LAI/TRL, regression and correlation analyses were performed using PROC REG and PROC CORR in SAS v. 9.4 statistical software (SAS Institute, 2013). The last measurement dates at approximately 75 DAP for both LAI and TRL were used to examine the relationship of these variables with pod yield. These relationships were separated by year and examined across all genotype and irrigation treatments.

Results

Yearly precipitation and irrigation: The annual precipitation between the two years was quite variable, with greater rates occurring during the 2015 growing season (834 mm) compared to the 2016 season (634 mm) (Figure 3.1). This resulted in substantial differences in the relative proportion of water received via irrigation and precipitation. The irrigation: precipitation ratio in 2015 and 2016 was 7 and 30%, respectively, in the 100% irrigation treatment (Table 3.1). The distribution pattern of precipitation also differed between the two years; for example, during the first 50 days after planting, approximately 125 mm more rainfall was received in 2016 compared to 2015. This difference is mostly attributed to one large precipitation event of 91 mm occurring on 5 June 2016. However, in both years soil water conditions required three irrigation treatments during the first 50 DAP. Precipitation following the first 50 DAP was substantial in 2015 and no additional irrigation was required (Figure 3.2). In contrast, very little precipitation was received from 50-95 DAP during the 2016 growing season, resulting in the need to apply seven irrigation treatments during this mid-season period.

Total Root Length: Differences in TRL growth were observed among the peanut genotypes by the first measurement day at 21 DAP (Table 3.2; Figure 3.3). Greater root growth occurred in COC 041 when compared to genotypes of *hypogaea* descent. At 28 DAP, greater separation in TRL occurred and the differences among genotypes continued to increase. Root growth rate appeared to slow for most genotypes by 35 DAP, with maximum TRL achieved by the last measurement date at 75 DAP. Total root length measurements after 35 DAP showed delineations between the two peanut market types: TRL was similar between the two Valencia market types; however both were greater than the two runner market types. Differences in TRL growth also existed

between the two runner market types with TUFRunner™ '511' having greater TRL growth than FloRun™ '107' (Figure 3.3).

Season-long patterns of TRL also varied among peanut genotypes by soil depth (Table 3.3; Figure 3.4). From 0-20 cm soil depth, COC 041 had a greater TRL than genotypes of *hypogaea* descent across all measurement dates; while at the 20-40 cm depth, TRL was similar among genotypes, with the exception of FloRun™ '107', which exhibited a shorter TRL than the other genotypes. At 40-60 cm depth, COC 041 and NMVC had similar TRL which was longer than the TRL of FloRun™ '107' and TUFRunner™ '511', both of which had similar TRL at this depth zone. At 60-80 cm of soil depth, all genotypes had similar TRL until 35 DAP. The final TRL measurement at 75 DAP at this depth range was similar to the trends in the 40-60 cm depth: Valencia market type cultivars had longer TRL than the runner market types, and genotypes within each market type were similar. Although these differences were observed for TRL, no significant differences in TSA where observed when making similar genotypic comparisons across the 20 cm soil depth increments over the growing season (Table 3.3; Figure 3.5). This result indicates that the TRL of genotypes of *hypogaea* descent generally have fewer large diameter roots, thus contributing to an overall similar amount of TSA for the genotypes.

Leaf Area Index: The results in both years show that early LAI development was not impacted by irrigation, but by 64 DAP, the 100% irrigation treatment had larger LAI than the RF (Table 3.2; Figure 3.6a). By 77 DAP, the 60%PA had achieved greater LAI than the RF treatment as well. Genotypic differences in LAI emerged by 77 DAP, a pattern that was consistent between years (Figure 3.6b). At 77 DAP, both runner market

types, FloRun^{TM} '107' and TUFRunner^{TM} '511', had similar LAI when compared to each other, and both had greater LAI than the two Valencia market types, NMVC and COC 041, both of which had similar LAI values.

Pod Yields: Irrigation treatments ultimately influenced peanut pod yields, a response that was similar across growing seasons and genotypes (Table 3.4). Treatments which received irrigation were statistically similar to one another, although all had greater yields than the RF treatment (Table 3.5). Irrigation treatments of 100%, 60%PA, and 60% had higher pod yields relative to the RF control of 23, 15, and 17%, respectively. Year also had a significant influence on genotypic pod yield (Table 3.6). In both years, yield followed an expectation of greater levels for the longer maturing runner market types than for the Valencia's. In 2016, even the runner market type yield differed between the two representative genotypes, with pod yield of FloRun^{TM} '107' being 672 kg ha^{-1} greater than TUFRunner^{TM} '511'. Yields were significantly reduced among all genotypes in 2016, a likely impact of less mid-season precipitation, resulting in yield declines, when comparing 2016 to 2015, of 21, 30, 68, and 59% for FloRun^{TM} '107', TUFRunner^{TM} '511', NMVC, and COC 041, respectively.

Morphological Partitioning and Pod Yields: Leaf area index was a significant predictor of pod yield across all genotypes and irrigation treatments in both years (Figure 3.7). However, variability in the predictive strength of this relationship was observed between the two growing seasons, with R^2 values of 0.52 and 0.15 in 2015 and 2016, respectively. This positive relationship had regression equations of yield = -241 + 946*LAI ($P < 0.0001$) and yield = -281 + 704*LAI ($P = 0.0015$) in 2015 and 2016, respectively. There was also a relationship between yield and TRL, showing R^2 values

of 0.15 and 0.21. This relationship was negative with regression equations of yield = 4398 – 1.10*TRL (P = 0.0015) and yield = 3422 – 1.37*TRL (P = 0.0002) in both 2015 and 2016, respectively (Figure 3.8).

Discussion

Reports have documented that the introduction of procumbent canopy growth habits of *hypogaea* over erect canopy growth habits of *fastigiata* descent have increased HI (Haro et al., 2013). Furthermore, Haro et al. (2017) documented that increases in HI were supported by increases in LAI, radiation intercepted by the crop, and a longer crop growing cycle. Greater LAI of genotypes with procumbent growth habits was also observed in this study. This study further advances the growth habit knowledge base of these two peanut subspecies by quantifying both above growth and below ground traits. Genotypic RSA differences were observed between the disparate subspecies indicating that their belowground traits are inherently different. Specifically, genotypes of *fastigiata* descent had greater root growth following 35 DAP which lasted until the final root measurement at 75 DAP. Additionally, the greater amount of TRL with genotypes of *fastigiata* descent occurred primarily at deeper soil depths ranging from 40-60 and 60-80 cm. Genotypic differences within subspecies *hypogaea* were observed in TRL (TUFRunner™ '511' > FloRun™ '107') when summed across the entire mini-rhizotron area. However, TSA was similar among all genotypes indicating two different RSAs among the two subspecies. These architectures include subspecies *fastigiata* having possibly greater TRL as a result of an increased number of lateral roots, whereas genotypes of *hypogaea* have fewer, more coarse roots. Genotypic differences in TRL within *hypogaea* could also be a result of increased genetic variation within this subspecies due to more extensive breeding and selection.

Both TRL and LAI observed on the final measurement date were correlated with pod yields. In both years, there was a significant weak negative relationship between pod yield and TRL. This indicates that part of the increased HI of the *hypogaea* subspecies, and selection for high yielding cultivars, may have been achieved by partitioning less carbon to root systems. Studies comparing wheat landraces to modern cultivars, which have been extensively selected for yield, have observed decreases in root biomass of the latter (Waines and Ehdaie, 2007). A positive relationship between pod yield and LAI was also observed in both years of this study. However, the relationship was much stronger (r=0.72) in 2015, whereas this relationship was weak (r=0.39) in 2016. A possible explanation for this is the variability in yearly environmental conditions. The 2015 season was characterized as relatively wet, with no irrigation applied after 47 DAP. Under non-water deficit conditions, biomass production is predominantly influenced by the fraction of intercepted photosynthetically active radiation (iPAR) and the efficiency of conversion of that solar energy into chemical energy (Charles-Edwards, 1982; Monteith, 1977). The interception of iPAR is largely a function of LAI and canopy architecture (Haro et al., 2017). However, under water deficit conditions the major limitation on biomass production is not iPAR but the lack of CO_2 substrate stemming from stomatal closure (de Wit, 1958; Donald and Hamblin, 1976; Blum, 2009).

The yearly precipitation patterns influenced water deficit stress severity and ultimately impacted genotypic pod yields. When averaged across all genotypes, yield showed a 39% reduction in 2016 compared to 2015. This is likely due to the more severe water deficit conditions in 2016. Additionally, a year x genotype interaction

provides evidence of differential pod yield susceptibility to water stress conditions. When comparing pod yields of 2016 to 2015, genotypes of subspecies *fastigiata* had yield declines of 64% when compared to a 25% pod yield reduction with genotypes of *hypogaea* descent. Genotypes of *fastigiata* descent may have greater susceptibility to pod yield reductions when subjected to more severe water deficit conditions. Interestingly, these genotypes were those which had the largest amount of TRL growth at deeper soil depths (40-60 and 60-80 cm). This was surprising and contrary to other studies that examine root traits in relation to drought stress. For example, Koolachart et al. (2013) examined the impacts of terminal water stress on peanut genotypes and demonstrated that peanut genotypes which maintain greater pod yields under water deficit stress are those which have more roots located deeper in the soil profile, and the ability to maintain transpiration rates. Other studies have demonstrated large variation among peanut genotypes in the fraction of transpirable water content when transpiration rates begin to decline due to soil drying (Devi et al., 2009). Therefore, it is plausible that, despite having greater amounts of TRL deeper in the soil profile, genotypes of *fastigiata* descent may have a greater sensitivity to soil drying leading to fast stomatal closure, thus necessarily limiting their pod yield potential under water deficit conditions. Furthermore, the possible fewer number of coarse lateral roots with subspecies *hypogaea* could be a more efficient strategy for maintaining water uptake under soil drying conditions in this environment. Large diameter roots may allow for improved root to soil water contact and hydraulic conductance under soil drying conditions in the porous sandy soils of this study.

The sensitivity of above and belowground traits to change in soil hydrological conditions was further evaluated in this study by examining these responses across several of irrigation management strategies. Beginning at 64 DAP, a general trend of reductions in LAI with decreasing amounts of applied irrigation water became evident. However, irrigation had no influence on RSA development, suggesting that leaf area development is more sensitive to water stress conditions than root growth. The sensitivity of leaf growth rate and an ability of roots to grow despite water stress has been documented (Boyer, 1968; Westgate and Boyer, 1985). Furthermore, Sharp et al. (1988) showed that, although reductions in leaf and root elongation rates can occur at the onset of soil drying in a controlled environment, continued soil drying reduced elongation of leaf tissues more than root tissues. Our results confirm the greater sensitivity of peanut canopy growth versus root growth to water deficit stress. This research also supports that, despite the inherent diversity among root and canopy morphology of these peanut subspecies, differential genotypic root and canopy responses may not be evident at the levels and duration of water stress conditions which typically occur in a humid climate.

A critical factor influencing root growth responses to irrigation management is the severity of water stress deficit at particular times in phenological development. The lack of root response to irrigation management factors in the present study could be due to similar amounts of irrigation applied during the first 50 DAP in both years; the period when peanut root development was most rapid and the overall seasonal architectural pattern is established. In both years, three irrigation treatments were applied during this time period which is relatively little water in proportion to the amount of precipitation

received. This could have contributed to the contrasting results of the current study with those observed by Rowland et al. (2012), where reduced irrigation early in the growing season resulted in increased root growth deeper into the soil profile. The arid climate where this study was conducted required a greater proportion of irrigation to precipitation. In both years of the current study, the three irrigation treatments prior to 50 DAP were applied during a short period of time resulting in the stress severity and duration being fairly mild or moderate. Plants which undergo moderate levels of water stress have been reported to have little difference in growth response compared to plants which have had no water stress (Padilla et al., 2009). A meta-analysis conducted by Poorter et al. (2012) demonstrated that a reduction in plant biomass and an increase in root mass fraction are often attributed to more severe water stress of longer durations. The authors hypothesized this to be an important adaptation of plants, because rapid alteration of growth partitioning may result in growth habits that are sub-optimal after the alleviation of water stress. This is applicable to the irrigated humid peanut production environment of this study where moderate intermittent water stress is often relieved by in-season rainfall events. Therefore, rapid partitioning of carbon for deeper roots alone could be costly and may be less able to acquire surficial soil water.

Conclusion

This research demonstrates that different inherent above and belowground traits exist among peanut genotypes of *fastigiata* and *hypogaea* descent, giving further insight into their whole plant carbon partitioning and adaptability to varying soil hydrological conditions. It has been documented that the greater yield potential of *hypogaea* subspecies is largely due to an increased crop cycle duration, and improvement of canopy dynamics to support greater biomass production and HI. This research also

concludes that part of the greater above ground biomass production with *hypogaea* subspecies may be due reduced amounts of TRL when compared to genotypes of *fastigiata* descent. Consequently, it could be hypothesized that decreases in TRL, particularly deeper in the soil profile, would result in the *hypogaea* subspecies to be less water deficit stress resilient. However, the results of this research demonstrate the contrary, at least to the level of water deficits experienced, where the pod yield reduction under increasing water deficit stress was greater in the *fastigiata* subspecies. This could be due to varying water acquisition efficiencies among the contrasting root architectures of the two subspecies. That is, fewer coarse roots may be more effective at water acquisition in a humid environment, particularly in soils possessing a high sand content, low water holding capacity, and high frequency surficial water inputs. However, it is important to note that greater water acquisition of a particular RSA may not result in greater water utilization of a crop genotype which has high stomatal sensitivity to soil drying. Future research which links peanut phenotypic data with crop water use function will provide greater understanding of genotypic variation in water acquisition and utilization. This would likely benefit selection methods for improving agronomic water deficit stress resiliency.

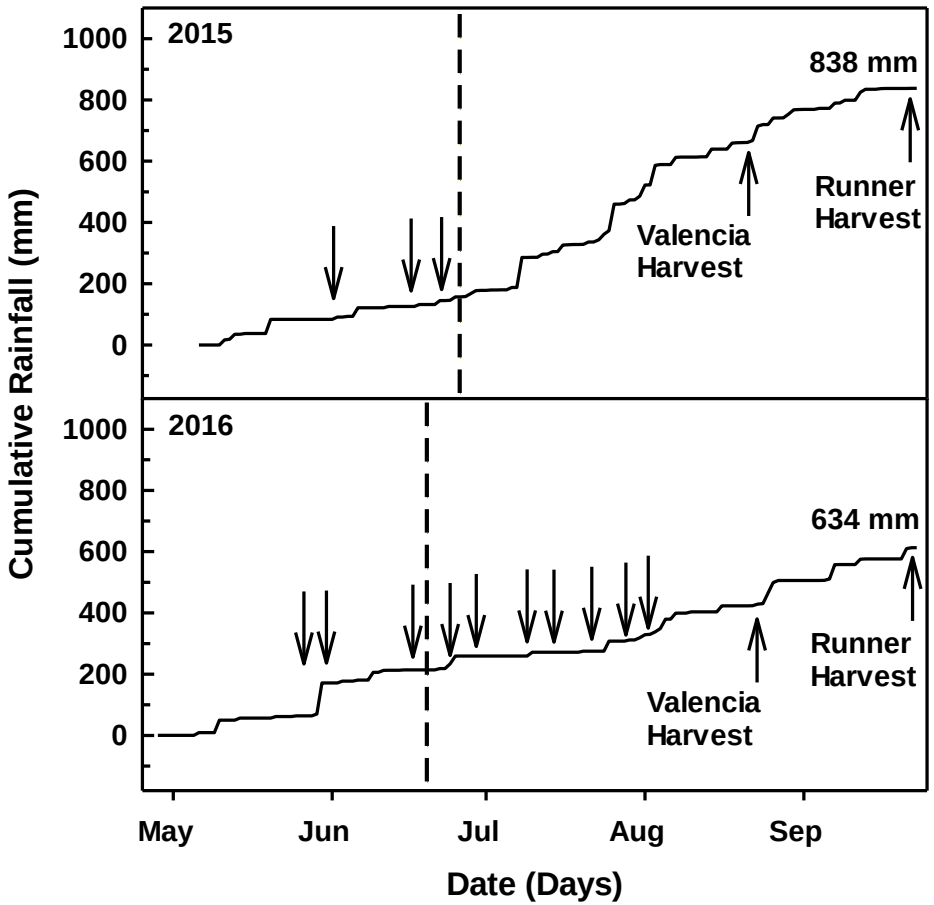

Figure 3-1. Cumulative precipitation from peanut planting to harvest in 2015 and 2016 at the Plant Science Research and Education Unit (PSREU) near Citra, FL. Vertical arrows pointing downward indicate when irrigation was applied. Dashed line represents 50 DAP when 60%PA irrigation treatment was similar to the 100%. Vertical arrows pointing up represent our two harvest dates for each peanut market type in both years (Abbreviations: 100%, 1.9 cm per application; 60%PA, 1.1 cm per application until 50 DAP, followed by 1.9 cm per application for the remainder of the growing season).

Table 3-1. Amounts of water received through irrigation, irrigation plus precipitation, and the irrigation to precipitation ratio in 2015 and 2016 for each irrigation treatment at the PSREU near Citra, FL.

	Irrigation Water Applied		Total Water Received		Irrigation: Precipitation	
	2015	2016	2015	2016	2015	2016
Irrigation Treatment	---------- mm ----------				---------- % ---------	
100%	57	191	894	825	7	30
60%PA	34	168	871	802	4	27
60%	34	114	871	748	4	18
Rainfed	0	0	837	634	0	0

† Abbreviations: 100%, 1.9 cm per application; 60%PA, 1.1 cm per application until 50 DAP, followed by 1.9 cm per application for the remainder of the growing season; 60%, 1.1 cm per application for the entire growing season; RF, rainfed control.

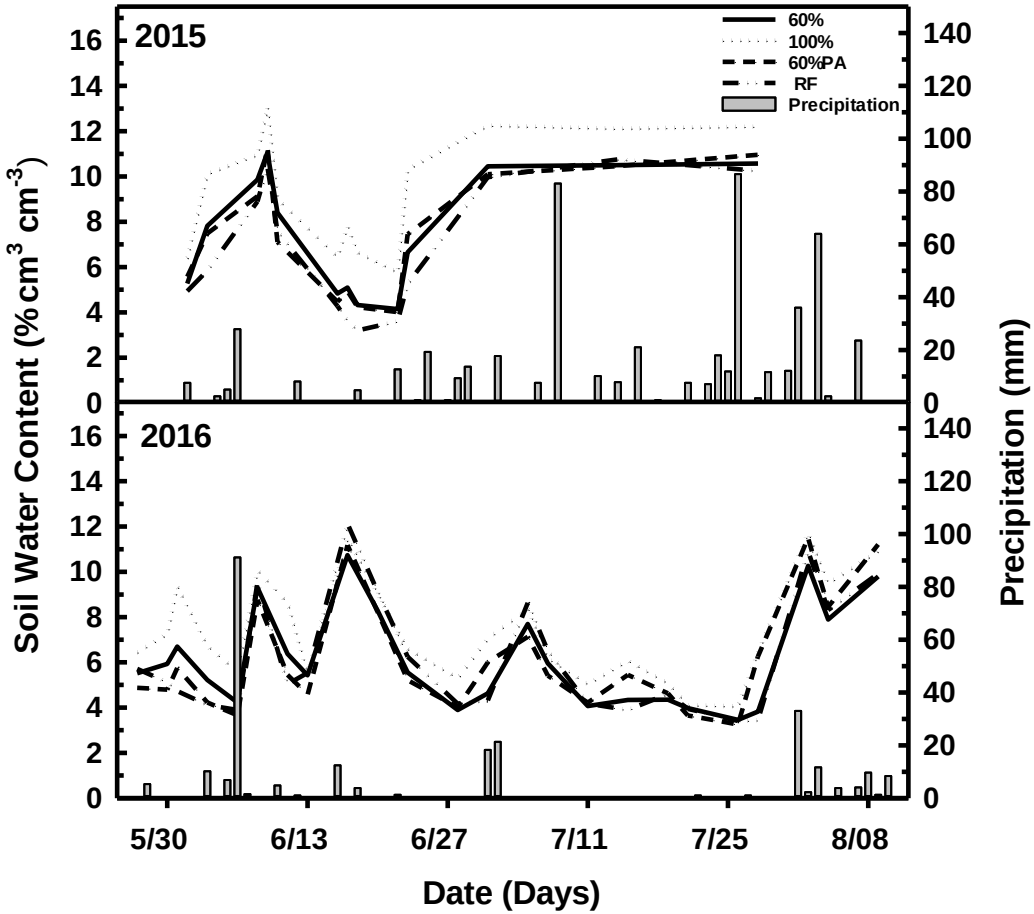

Figure 3-2. Volumetric soil water content (left axis) and daily precipitation (right axis) during time periods when irrigation was applied in 2015 and 2016 at the PSREU near Citra, FL. (Abbreviations: 100%, 1.9 cm per application; 60%PA, 1.1 cm per application until 50 DAP, followed by 1.9 cm per application for the remainder of the growing season).

Table 3-2. Analysis of variance (ANOVA) for response variables of TRL, TSA, and LAI. Root parameter data was summed across soil depths prior to running ANOVA.

Effect	Total Root Length	Total Surface Area	Leaf Area Index
year	0.3323	0.5180	0.0145
IR	0.6748	0.7169	0.1053
year*IR	0.8525	0.8403	0.9444
GENO	<0.0001	0.1122	0.0007
year*GENO	0.9164	0.7960	0.0007
IR*GENO	0.2123	0.1267	0.1236
year*IR*GENO	0.8085	0.9005	0.6393
TM	<0.0001	<0.0001	<0.0001
year*TM	<0.0001	<0.0001	0.3319
IR*TM	0.4050	0.1295	0.0006
year*IR*TM	0.1266	0.1424	0.3961
GENO*TM	0.0014	0.4635	<0.0001
year*GENO*TM	0.7099	0.6589	0.2444
IR*GENO*TM	0.9508	0.9731	0.9908
year*IR*GENO*TM	0.8519	0.8154	0.8029

†Abbreviations: year, growing seasons of 2015 and 2016 at the PSREU; IR, irrigation treatment; GENO, genotype treatment; TM, time repeated measures within each growing season.

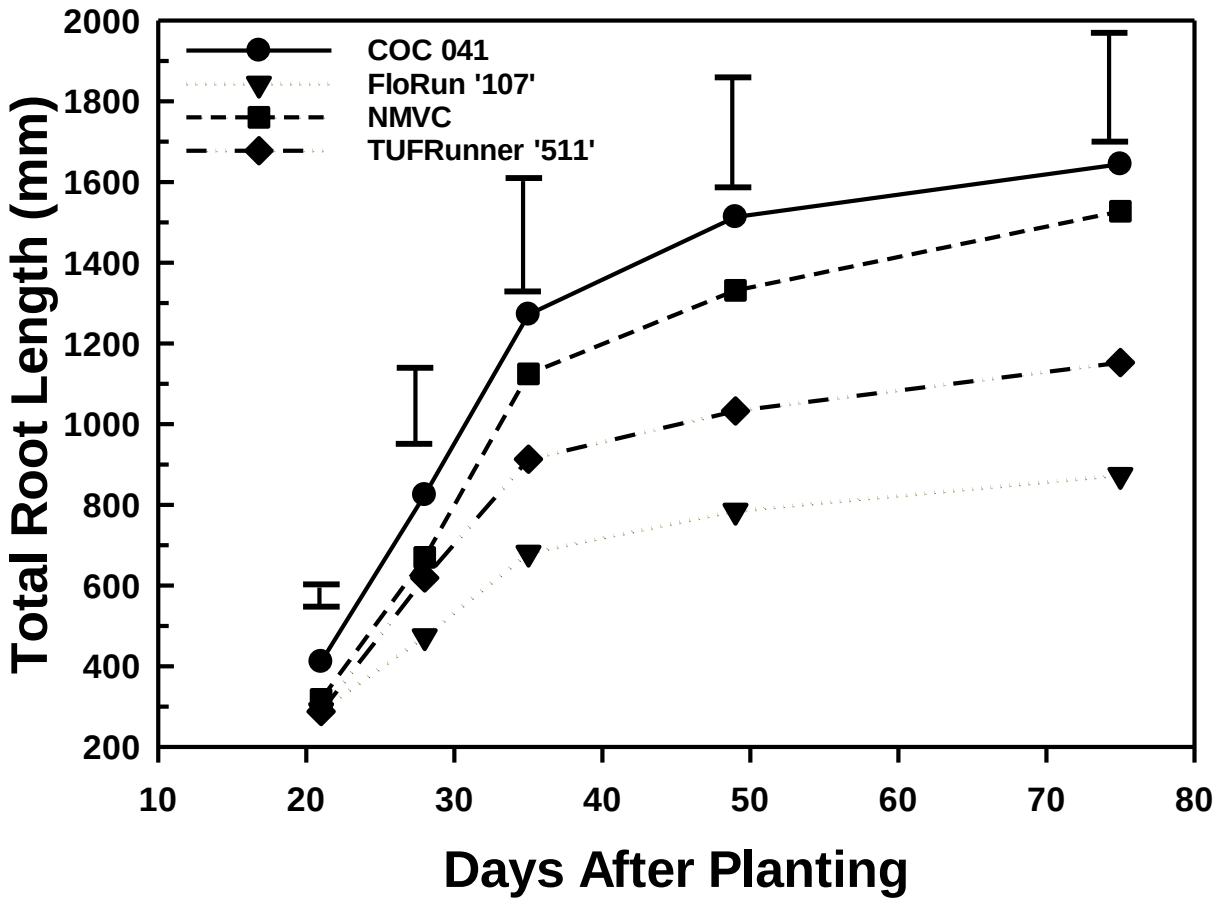

Figure 3-3. Average total genotypic root length growth at 21, 28, 35, 49, and 75 DAP at the PSREU. Data is averaged across irrigation treatments and year. Error bars are Fisher's Protected Least Significant Difference at $P < 0.05$. Total root length is the sum of root lengths across the entire mini-rhizotron tube to a soil depth of 80 cm (Abbreviations: NMVC, New Mexico Valencia C).

Table 3-3. Analysis of variance (ANOVA) for response variables of TRL and TSA. Response variables were summed across 20 cm soil depth increments prior to running ANOVA.

Effect	Total Root Length	Total Surface Area
Year	0.1160	0.9928
IR	0.5521	0.3708
Year*IR	0.7393	0.9243
GENO	<0.0001	0.0247
Year*GENO	0.8816	0.1852
IR*GENO	0.0123	0.3944
Year*IR*GENO	0.4221	0.0173
Depth	<0.0001	<0.0001
Year*Depth	0.0447	0.2308
IR*Depth	0.4982	0.9184
Year*IR*Depth	0.9885	0.2108
GENO*Depth	0.8995	0.9552
Year*GENO*Depth	0.3072	0.9196
IR*GENO*Depth	0.8337	0.8977
Year*IR*GENO*Depth	0.9851	0.2329
TM	<0.0001	<0.0001
Year*TM	<0.0001	0.0006
IR*TM	0.0689	0.0149
Year*IR*TM	0.0522	0.0583
GENO*TM	<0.0001	0.0006
Year*GENO*TM	0.7531	0.4423
IR*GENO*TM	0.4807	<0.0001
Year*IR*GENO*TM	0.2078	0.3791
Depth*TM	<0.0001	<0.0001
Year*Depth*TM	<0.0001	0.0202
IR*Depth*TM	0.3291	0.1173
Year*IR*Depth*TM	0.5564	0.5885
GENO*Depth*TM	0.0345	0.9829
Year*GENO*Depth*TM	0.6818	0.8541
IR*GENO*Depth*TM	0.9703	0.1983
Year*IR*GENO*Depth*TM	0.4530	0.9237

†Abbreviations: year, growing seasons of 2015 and 2016 at PSREU; IR, irrigation treatment; GENO, genotype treatment; TM, time repeated measures within each growing season; Depth, sum of root parameters across 20 cm depth increments.

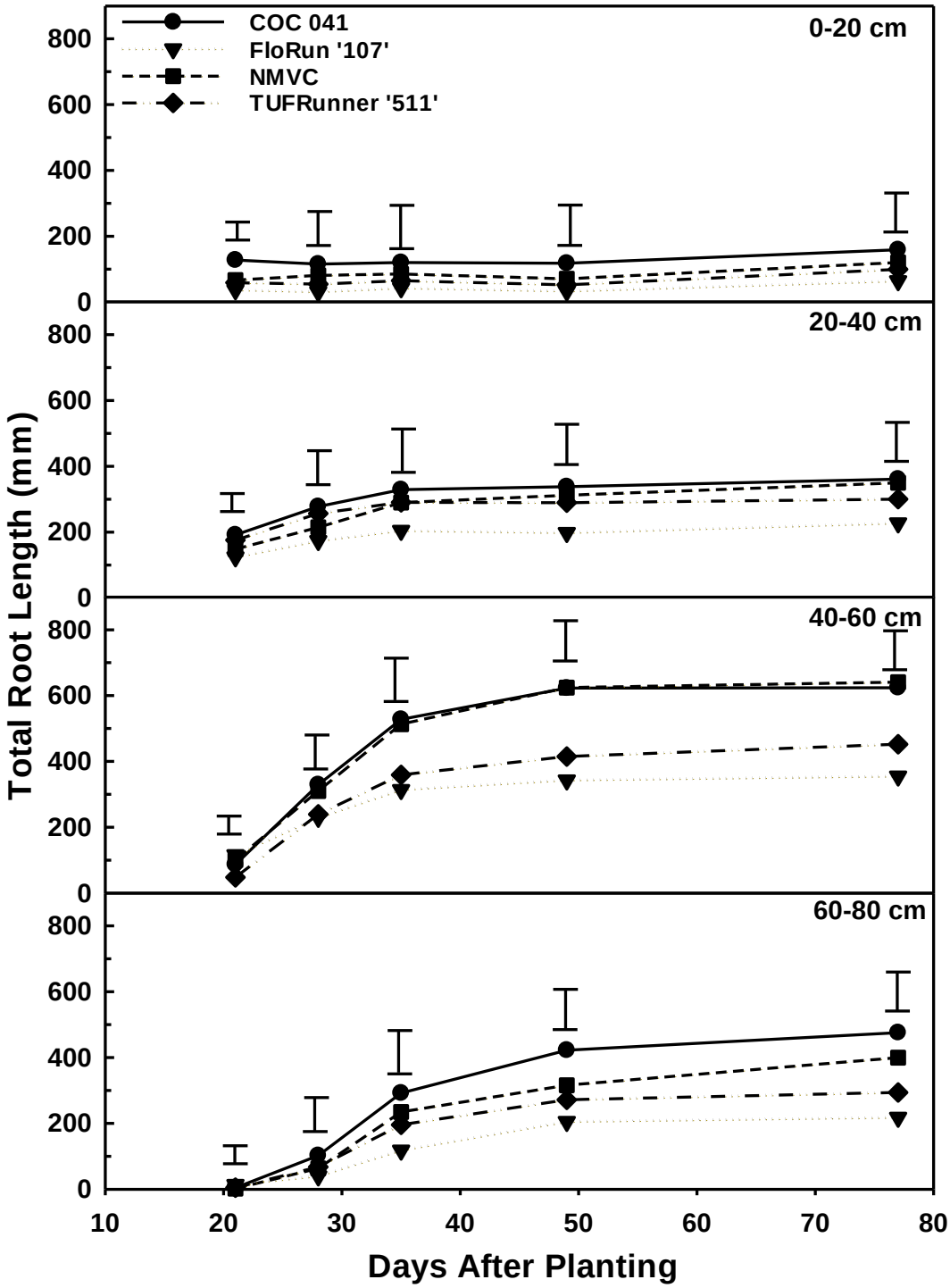

Figure 3-4. Average genotypic root length summed across 20 cm depths at 21, 28, 35, 49, and 75 DAP at the PSREU. Error bars are Fisher's Protected Least Significant Difference at $P < 0.05$. Data is the averaged across irrigation treatments and years (Abbreviations: NMVC, New Mexico Valencia C).

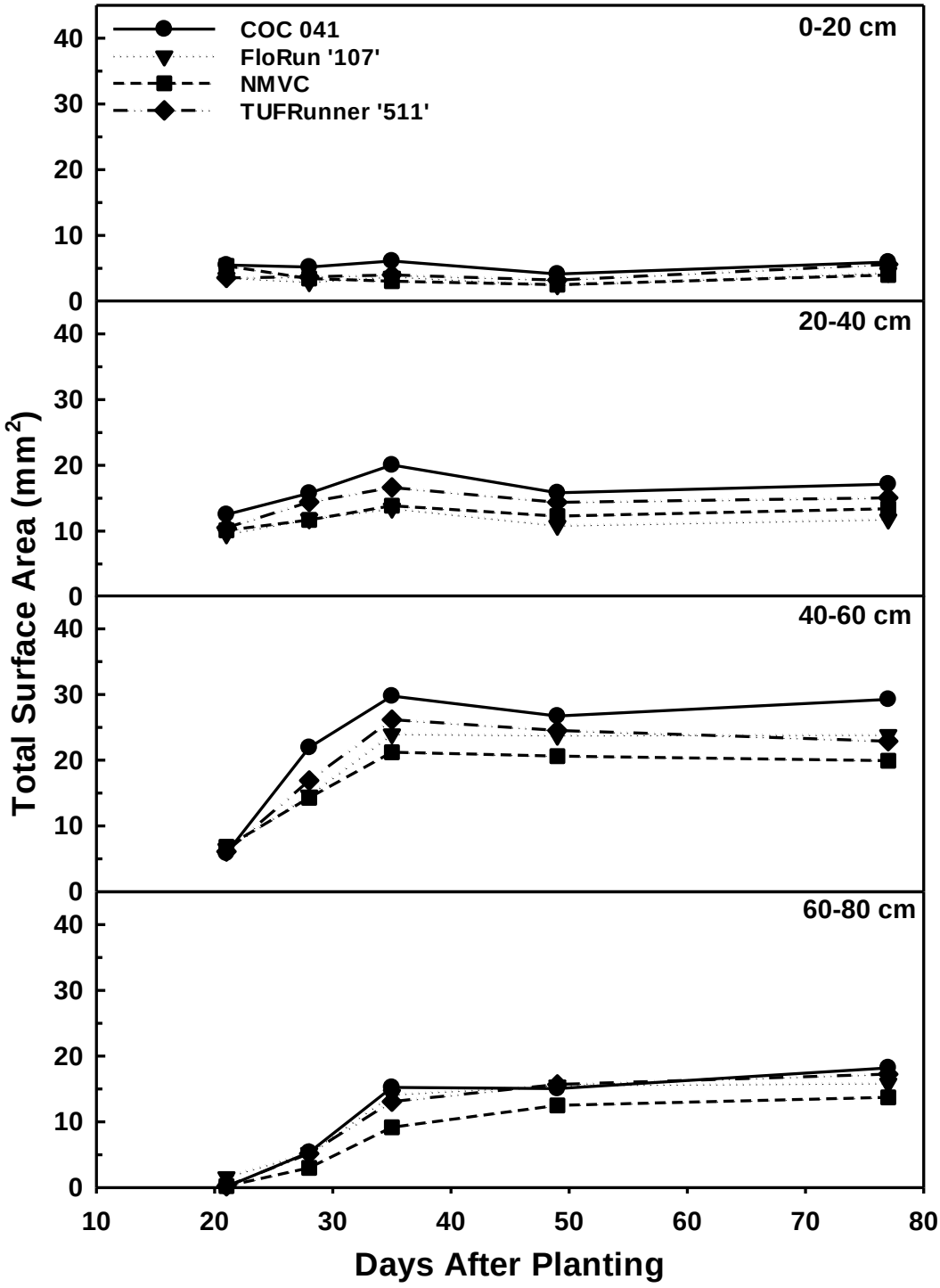

Figure 3-5. Average genotypic root surface area summed across 20 cm depths at 21, 28, 35, 49, and 75 DAP at the PSREU. No multiple comparisons error bars are displayed because F-test results had a P > 0.05. Data is the averaged across all irrigation treatments and years. (Abbreviations: NMVC, New Mexico Valencia C).

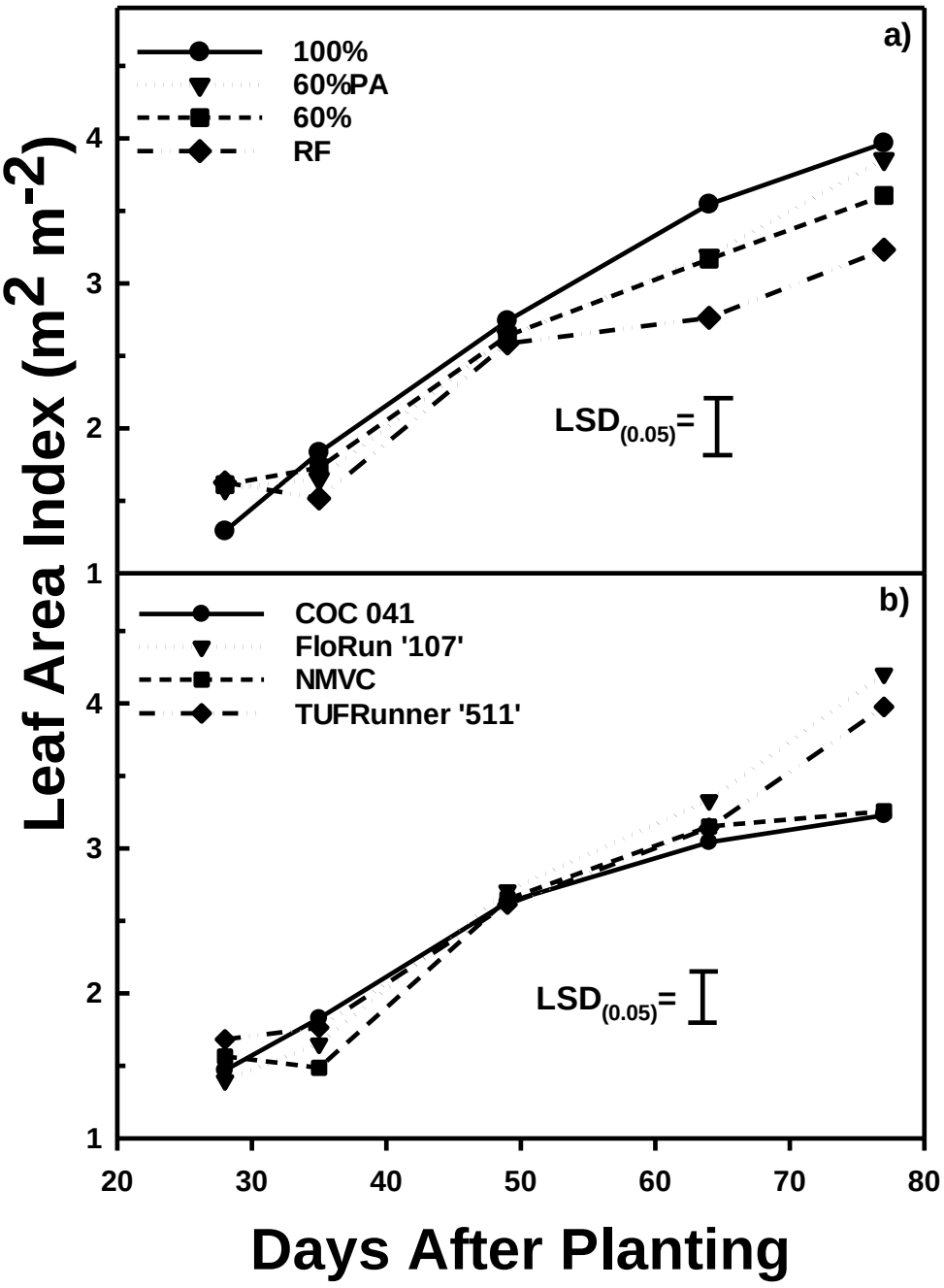

Figure 3-6. A) Average leaf area index (LAI) at 28, 35, 49, 64, and 77 DAP for each irrigation treatment and B) genotype at the PSREU. Data is averaged across years for graphs A & B (Abbreviations: 100%, 1.9 cm per application; 60%PA, 1.1 cm per application until 50 DAP, followed by 1.9 cm per application for the remainder of the growing season; 60%, 1.1 cm per application for the entire growing season; RF, rainfed control; LSD, Fisher's Protected Least Significant Difference at $P < 0.05$).

Table 3-4. Analysis of variance (ANOVA) for pod yield response variables at the PSREU near Citra, FL.

Effect	Pod Yield
year	0.0022
IR	0.0054
year*IR	0.7077
Geno	<0.0001
year*Geno	0.0301
IR*Geno	0.1581
year*IR*Geno	0.2575

†Abbreviations: year, growing seasons of 2015 and 2016; IR, irrigation treatment; GENO, genotype treatment; TM, time repeated measures within each growing season.

Table 3-5. Average pod yields for each irrigation treatment at the PSREU near Citra, FL. Yields are averaged across peanut genotypes and years.

Pod Yields	
Irrigation Treatment	Pod Yield
	------- kg ha^{-1} -------
100%	3103
60%PA	2801
60%	2886
RF	2382
$LSD_{(0.05)}$	369

† Abbreviations: 100%, 1.9 cm per application; 60%PA, 1.1 cm per application until 50 DAP, followed by 1.9 cm per application for the remainder of the growing season; 60%, 1.1 cm per application for the entire growing season; RF, rainfed control.

Table 3-6. Average pod yields for each genotype and growing season at the PSREU near Citra, FL. The percent difference is the percentage of yield decline from 2015 to 2016.

Peanut Genotypes	Peanut Pod Yields 2015	2016	Percent Difference
	---------- kg ha^{-1} ----------		--------- % -------
FloRun™ '107'	4718	3731	21
TUFRunner™ '511'	4373	3059	30
NMVC	2532	815	68
COC 041	2227	907	59
$LSD_{(0.05)}$	---------- 609 ----------		-

†Abbreviations: NMVC, New Mexico Valencia C; LSD, Fisher's Protected Least Significant Difference at $P < 0.05$.

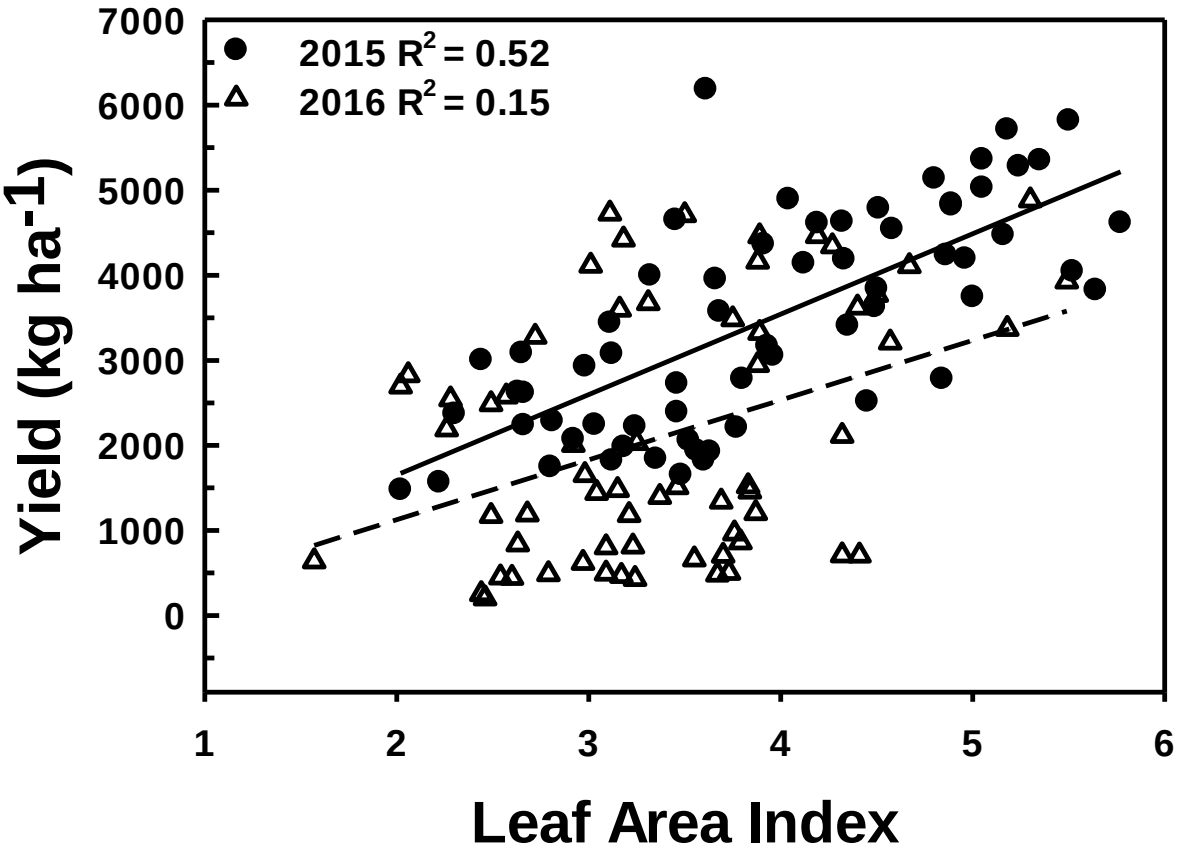

Figure 3-7. Relationship between peanut pod yield and maximum leaf area index achieved for the 2015 and 2016 growing season at the PSREU near Citra, FL. Individual data points represent all genotypes and irrigation treatments for each year.

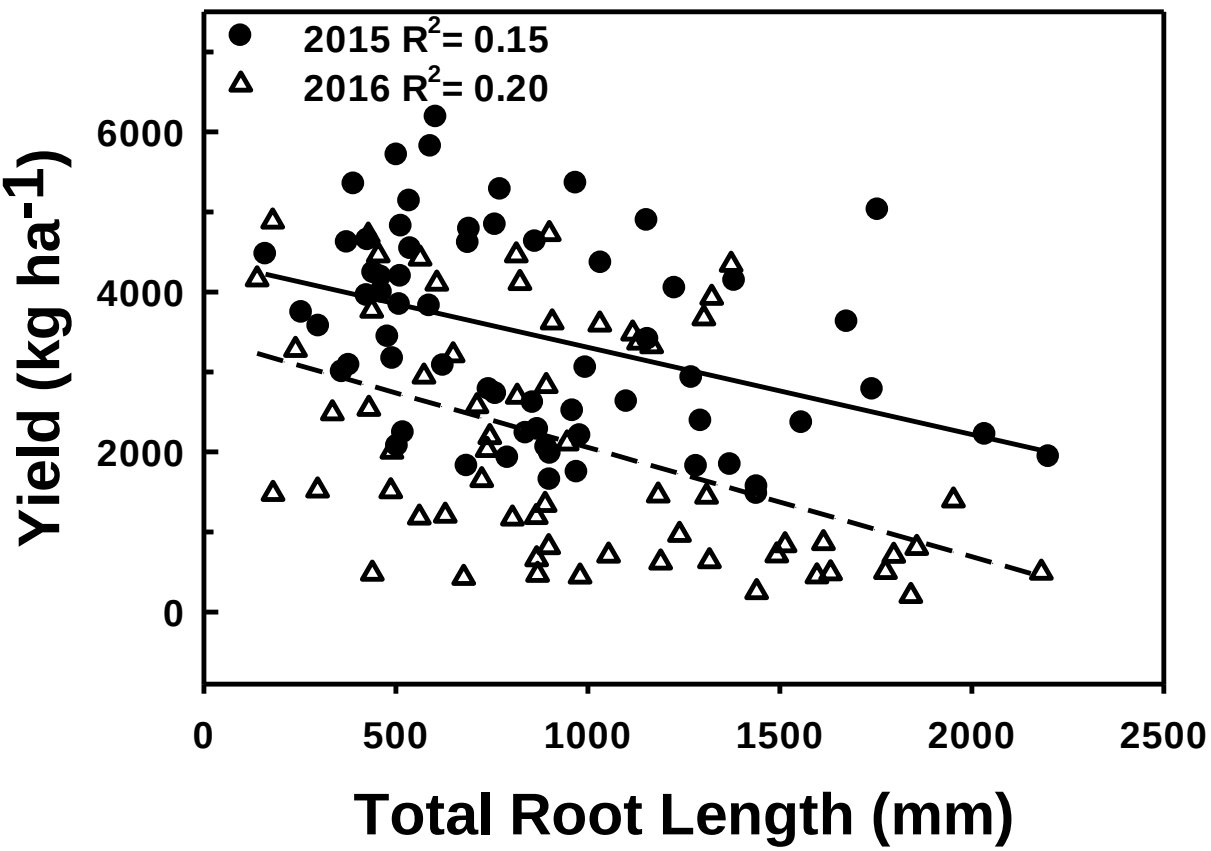

Figure 3-8. Relationship among pod yield and maximum total root length achieved for the 2015 and 2016 growing season at the PSREU near Citra, FL. Individual data points represent all genotypes and irrigation treatments for each year.

CHAPTER 4
COORDINATED ADAPTIVE PHENOTYPING FOR IMPROVING SOIL WATER ACQUISITION AND UTILIZATION

Introduction

Drought stress is responsible for more crop loss than any other abiotic or biotic stress, thus making tolerance to water scarcity a critical goal in many breeding programs (Borém et al., 2012). But progress has been slow, primarily due to both the complexity of conditions that have been categorized under the general term “drought” (Mwadzingeni et al., 2016), and because of the breadth of individual plant processes and traits which contribute to “tolerance” to water scarcity (Cattivelli et al., 2008; Sinclair, 2011; Turner et al., 2014). In addition, the concept of successful drought tolerance in crop species diverges from many established paradigms regarding plant drought tolerance in general. This is because operationally, drought tolerance has been repeatedly defined as the ability to survive under research scenarios that compare well-watered conditions to severe water reductions. However, this definition and method of evaluation is not relevant to crop species because they must do more than survive – they must continue to yield under water scarcity, necessarily making genetic and physiological mechanisms of yield maintenance inherently different than those of survival (Skirycz et al., 2011). All these factors have led to the lack of true breakthroughs in solving the issue of breeding for crop drought tolerance.

To successfully phenotype crop genotypes for water deficit stress tolerance, it is important to examine responses in both above- and below-ground traits, but perhaps more importantly, to assess their coordinated functioning. One of the most impactful aboveground traits which influences crop yield under water deficit stress is the degree of transpiration reduction which occurs under water deficit stress (de Wit, 1958;

Passioura, 1996; Blum, 2009). The ability to maintain transpiration under dehydrating conditions allows for continued assimilation of CO_2 during photosynthesis, which is required for growth processes. Variability exists among peanut genotypes for the sensitivity of transpiration reductions under soil drying (Devi et al., 2009). Utilizing the relationship between stomatal closure and carbon isotope discrimination, Hubick et al. (1988) found a broad sense heritability of 81% in carbon isotope discrimination, suggesting that transpiration use efficiency (a trait influenced by stomatal closure) is a heritable trait which can be utilized in breeding programs. These results indicate that selection and breeding for stomatal sensitivity to water deficit stress can be used to mitigate yield loss, or possibly conserve water to avoid lethal levels of crop desiccation.

For critical belowground traits, root system architecture (RSA) encompasses a suite of traits which have the potential to provide genotypic water deficit stress tolerance. For peanut, genotypes with inherently deep root architectures and the ability to proliferate roots deep into the soil profile under dehydrating conditions have relatively greater pod yields under water stress conditions (Rucker et al., 1995; Songsri et al., 2008; Jongrungklang et al., 2011; Jongrungklang et al., 2012; Koolachart et al., 2013). But through the increased emphasis on examining roots traits in general, research is showing that an examination that focuses solely on root length and depth is not correctly predicting the complex response to soil drying and thus the overall tolerance to water deficits. Similarly for peanut, the relationship between extensive root architectures and dehydration tolerance is not always clear, including Ratnakumer and Vadez (2011) who reported a weak positive relationship between peanut root length density (RLD) and total water uptake under various water stress conditions. Therefore, concluding that

deep and extensive RSAs are the primary or only phenotypes appropriate for drought tolerance is misguided.

It is clear that individually, above- and belowground traits play varying roles in providing adaptability to various dehydration vulnerability scenarios (DVS). However, what may be even more critical is the integration of function across above- and belowground processes. This idea is central to succeeding at identifying water deficit stress tolerant phenotypes because the entire water conductivity system of a plant is driven by transpiration, through a coordinated process of root water uptake from the soil, transport in the plant vascular system, and eventual water loss at the leaf surface. For example, a major resistance to transpiration rates under soil drying conditions is stomatal limitation, which would diminish root water uptake, regardless of where roots are distributed in the soil profile. Therefore, a genotype with an extensive root system that is able to access deep water resources but closes stomates quickly in a water scarce condition will not be able to fully capitalize on its extensive root system. In fact, while sensitive stomatal closure under developing soil drying conditions combined with a deep RSA may be excellently adapted for *survival* under drought, we could define this genotype as being *maladapted* to a commercial production setting because rapid stomatal closure will not allow this genotype to efficiently utilize the energy invested into an extensive RSA, likely causing limits to yield potential. On the contrary, a shallow rooted genotype with delayed stomatal sensitivity to soil drying would result in relatively low energy investment to RSA and thus increased yield potential. However, this same genotype would likely require adequate and reliable water application. Overall, agronomic water stress tolerance cannot be attributed to a single above- or

belowground trait, but that a coordinated combination of above- and belowground traits is required to maintain yield under water stress conditions.

While coordination of above- and belowground traits is essential for determining water scarcity tolerance, the degree of variability in the coordination of stomatal aperture control coupled with RSA could be useful in determining specific adaptability to a particular (DVS). The first objective of this research was to: (i) develop an *in situ* approach to quantify the coupled influence of root system architecture (RSA), soil water depletion (SWD), and stomatal limitation on soil water acquisition distribution within the soil profile over a series of soil wetting and drying; and (ii) determine if genotypic RSA responses to differential early season water management has a lasting effect on mid-season soil water depletion zones and transpiration fluxes over a series of soil wetting and drying cycles. We hypothesized that under well-watered conditions with no stomatal limitation, soil water depletion would primarily occur at the soil depth with the greatest root density. Additionally, the degree of RSA plasticity to differential early season water management would vary among peanut genotypes, and would ultimately determine the level of genotypic water deficit stress tolerance.

Materials and Methods

Site Characterization: A field study was conducted during 2016 at the University of Florida's Plant Science Research and Education Unit in North-Central Florida (29° 24' 38" N, 82° 10' 12" W). The soil is classified as Candler sand (Hyperthermic, uncoated Lamellic Quartzipsamments). Soil preparation consisted of conventional tillage approximately five weeks before planting. Immediately prior to planting, the field was surface tilled using a field cultivator with S-tine sweeps. Planting was accomplished with a two-row Monsem® planter (Monosem Inc., Edwardsville, KS) at a seeding density for

all peanut genotypes of 20 seeds m^{-1} with a row spacing of 0.91 m; plots consisted of four rows 6.0 m long. Rhizobium inoculum (1.31 l ha^{-1} ; Advanced Biological Marketing, Van Wert, OH)), and azoxystrobin fungicide (0.21 kg a.i. ha^{-1}; Sygenta International AG, Basel, Switzerland) were applied in the seed furrow at planting. A soil test was conducted approximately six weeks prior to planting, and recommended nutrients were applied on the surface immediately after planting.

Experimental Design: The experimental design consisted of a split plot arrangement in a randomized complete block design with three replicates. The whole plots were irrigation treatments consisting of: 1) 1.9 cm per application for the entire season scheduled using tensiometers (100%); and 2) a primed acclimation (PA) treatment consisting of 1.1 cm of water per application for the first 50 days after planting (DAP) followed by 1.9 cm applications for the remainder of the season (60%PA), with all scheduling of irrigation events following the 100% treatment. Irrigation was scheduled when tensiometers placed in the 100% irrigation treatment of each replication reached an approximate average soil water tension of -25 kPa at a soil depth of 30 cm. Irrigation was applied using surface drip irrigation placed directly on the row with a 30 cm emitter spacing producing 3721 L hr^{-1} 100 m^{-1} at 69 kPa (JAIN Irrigation Inc, Haines City, FL). Sub-plots consisted of two genotypes, a Valencia genotype (*Arachis hypogaea* L. var. *fastigiata*) COC 041 (PI 493631), and a Runner (*Arachis hypogaea* L. var. *hypogaea*) commercial cultivar TUFRunner™ '511'.

Rainout shelters were used in this experiment to exclude rainfall from the plots to ensure differential irrigation treatments were precise and that a series of soil drying events at controlled levels could be applied during the peak growth period when in-

depth physiological characterization took place. These physiological measurements occurred from approximately 60-90 DAP. This time period was established for two primary reasons: 1) RSA in peanut is well established and at its peak extension by 60 DAP; and 2) this time period coincides with the phenological development period when peanut is most susceptible to pod yield loss due to water deficit stress. Testing during these dry-down periods allowed for a comparison of how early season irrigation management influenced RSA, and if the established RSA has lasting impacts on mid-season soil water acquisition.

Crop Measurements: Two mini-rhizotron tubes were installed in each plot directly into and parallel to the row at a 45° angle with the soil surface immediately after crop emergence using a hydraulic powered coring machine (Giddings Machine Company, Windsor, CO). At each measurement date, images were captured at 13.5 mm increments along the mini-rhizotron tubes using a BTC 100X video camera and BTC I-CAP image capture software (Bartz Technology Corporation, Carpinteria, CA). Total root length (TRL) and total root volume (TRV) analysis was conducted using WinRHIZO Tron software (Regent Instruments Inc., Quebec, Canada) by hand tracing of root segments within each image frame. Total root length on each image frame was computed by estimating the linear length of each root segment, while TRV was the product of total length and the cross sectional area ($CSA= \Pi r^2$) ($TRV=\Pi r^2 * TRL$) determined from the traced images. Root image sampling dates occurred at 60, 67, 75, 82, and 88 days after planting (DAP) to coincide with the dry-down periods.

Measurements of leaf level gas exchange were recorded using a LICOR-6400XT portable photosynthesis system at these same time periods coinciding with root

imaging, specifically at 61, 62, 68, 69, 75, 76, 82, 83, 89, 90, 95, and 96 DAP (LICOR, Lincoln, Nebraska). Two separate leaf measurements were recorded on the upper most mature leaf (second nodal position) located on the terminal branch, using one leaflet from each leaf in order to fill the measurement chamber. Leaves were selected from plant canopies whose RSA was being quantified using the mini-rhizotrons. Chamber conditions were consistent across sampling date, including a block temperature of 30° C, CO_2 concentration of 400 ppm, and a relative humidity maintained from 65-75%. The LED light source was set to 1800 µmol m^{-2} s^{-1}. The chamber flow rate was set to 500 µmol s^{-1}.

Concomitant with root imaging and gas exchange measurements, VSWC was quantified using time domain reflectometry (TDR) with the TRIME-PICO IPH/T3 probe (IMKO, Ettlingen, Germany). The probe sensor head was factory calibrated for the mini-rhizotron tube cellulose acetate butyrate (CAB) material. This allowed for quantification of the VSWC around each mini-rhizotron tube. The probe head measures the average VSWC at approximately 15.5 cm increments. These measurements were recorded on the same days as the gas exchange measurements: 61, 62, 68, 69, 75, 76, 82, 83, 89, 90, 95, and 96 DAP. Soil water depletion was estimated by taking the difference between pre-dawn and pre-dusk for each quantified soil depth at 61, 68, 75, 82, 89, and 96 DAP.

Statistical Analysis: Statistical analysis was performed using SAS v. 9.4 statistical software (SAS Institute, 2013). The GLIMMIX procedure was used to compute analysis of variance (ANOVA). Random effects of rep, and rep crossed with irrigation were included in the model. A repeated measure ANOVA was performed on

measurements that were repeated over the growing season (TRL, TRV, SVWC, gas exchange, SWD), with a covariance matrix structure with the lowest Akaike Information Criterion (AIC). An autoregressive (ARH) covariance matrix structure was used for TRL and TRV measurements. A heterogeneous autoregressive (ARH1) covariance matrix structure was used for leaf level transpiration rates, SWD, and VSWC measurements. Normality and homogeneity were visually assessed by graphing the residual distribution, scatter plot of residuals, and Q-Q plot of residuals. The TRL and TRV traits were heteroscedastic, so a square root transformation was performed prior to running the repeated measures ANOVA. Data were pooled over experimental treatments when F-tests were not significant (specifically on irrigation treatments of SWD and transpiration). Multiple comparisons were carried out using Fisher's Protected Least Significant Difference (LSD) at the $P < 0.05$ probability level.

To assess the bi-variate relationships between transpiration fluxes, root parameters, and soil water depletion, regression analysis was performed using SAS JMP Pro v. 12.2.0 statistical software (SAS Institute, 2015). A quadratic regression model was fit to the relationship between the daily transpiration flux and total soil water depletion summed to a soil depth of 70 cm. A linear regression model was fit to the relationship between TRV with soil water depletion for each genotype. A spearman correlation analysis was performed assessing the relationship between root parameters and SWD for all quantified soil depths and measurement dates. A spearman correlation was also used to estimate the relationship between VSWC and transpiration for each genotype across all measurement dates. A linear regression model and Pearson

correlation analysis were performed examining the relationship between transpiration rates and pod yield

Results

Root System Architecture: The RSA varied among the two peanut genotypes in this study (Table 4.1). In general, genotype COC 041 had a greater TRL than TUFRunner^{TM} '511', consisting of more fine to medium diameter classes (Figure 4.1); TUFRunner^{TM} '511' had the majority of roots in diameter classes greater than 3.5 mm 10^{-1}. Genotypic RSA also interacted with soil depth. Both genotypes COC 041 and TUFRunner^{TM} '511' had similar TRL growth in the top 20 cm of soil depth (Figure 4.2a), while COC 041 had greater TRL at soil depths below 20 cm. Because it had more coarse roots, the TUFRunner^{TM} '511' TRV in the 0-20 cm of soil depth was greater than COC 041 (Table 4.1), but below 20 cm depth, the TRV was similar among the two genotypes (Figure 4.2 b).

Early season irrigation patterns influenced the genotypic RSA (Figure 4.3 a & b). The genotype COC 041 responded to the 60%PA treatment by having greater TRV at the deepest quantified soil depth range of 60-80 cm, while the TRV in the top 60 cm of soil was not affected for this genotype (Figure 4.3a). Similarly, early season irrigation treatments affected the RSA of TUFRunner^{TM} '511' plants, with the 100% irrigation treatment having greater TRV in the soil depth ranges of 0-20 and 20-40 cm when compared to the 60%PA irrigation treatment, with the TRV for this genotype reduced in the 100% irrigation treatment at 60-80 cm of soil.

Soil Water Depletion and Transpiration: The distribution of soil water depletion across the soil profile varied over the growing season and was dependent on the overall VSWC (Figure 4.4 a&b). Under high VSWC, the estimated percent of soil water

depletion occurring in the top 30 cm of soil was 86, 66, and 71% at 61, 75, and 89 DAP, respectively. As the soil dried, most soil water depletion occurred at the depth of 30-60 cm. On these dates of 68, 82, and 96 DAP, the estimated percent of soil water depletion at 30-60 cm was 59, 67, and 76%, respectively. Significant correlations were observed between all rooting parameters and SWD. The measurement dates of 75 and 82 DAP which had contrasting VSWC levels demonstrate the general trend between SWD and the rooting parameters that was observed (Tables 4.2 & 4.3). The slope direction of the relationship was dependent on VSWC, such that on days where VSWC was high, and root uptake was predominately in the top 30 cm of soil, the amount of SWD increased with decreasing amount of roots root (negative slope) (all parameters). A positive slope was observed between SWD and root parameters when VSWC was low and the predominant amount of SWD was at soil depths greater that 30 cm.

When examining transpiration rates on these same dates, TUFRunner^{TM} '511' had greater mid-morning transpiration rates than COC 041 on the majority of the measurement dates (Table 4.4; Figure 4.5). At 61 and 83 DAP; TUFRunner^{TM} '511' had greater mid-morning transpiration rates despite similar values of VSWC from 0-70 cm of soil. However, greater VSWC with TUFRunner^{TM} '511' was observed at 62, 68, 69, 82, 90, and 96 DAP indicating less soil water depletion with this genotype. On days following irrigation treatments (75, 76, and 89 DAP) when VSWC had increased to high levels the mid-morning transpiration rates among the two genotypes were similar. During soil dry-down periods (62, 68, 69, 82, 90, 96, and 97 DAP) greater mid-morning transpiration rates were observed in TUFRunner^{TM} '511', with the exception of 97 DAP when a severe level of water deficit occurred (VSWC= 2.7%). The correlation between

VSWV and transpiration showed a positive correlation for both genotypes (COC 041, r= 0.48; TUFRunner^{TM} '511', r=0.28) (Table 4.5).

When examining the relationship between the daily transpiration flux and total soil water depletion summed to a soil depth of 70 cm (Figure 4.6), moderately high coefficients of determination (R^2=0.61 and 0.67) were estimated for the genotypes COC 041 and TUFRunner^{TM} '511', respectively. The quadratic regression equation was TR= 1.102 + 0.911*SWD – 0.040*SWD^2 (P< 0.0001) for COC 041, and TR= 2.292 + 0.958*SWD – 0.044*SWD^2 (P< 0.0001) for TUFRunner^{TM} '511'. When examining the linear relationship between TRV and SWD, an R^2=0.17 (TRV= 74.16 + 3.527*SWD; P= 0.021) and 0.00 for genotypes COC 041 and TUFRunner^{TM}'511', respectively was observed. These results demonstrate that soil water depletion is more strongly related to daily transpiration flux than total root system size.

Discussion

To date, research interests focusing on examining variation in genotypic root architecture and plasticity has gained momentum due to its potential for identifying traits which could improve crop resilience to both impaired water and nutrient availability and ultimately contribute to resource use efficiency. However, very few of these studies attempt to quantify the relationship between root architecture and water uptake which leads to many assumed causal links regarding physical root presence and water uptake capacity. The lack of directly relating root presence with quantified measures of water uptake is due to many logistical and expense restraints which make this measurement challenging. Estimations of crop water use using weighing lysimeters and sap flow rates are not only expensive (reducing the number of genotypes in which crop water use can be estimated), but do not provide information on the distribution of soil water depletion

in relation to root water uptake. Utilizing soil water content sensors to examine soil water depletion can also be challenging if not properly paired with estimates of root architecture. Therefore, an objective of this study was to examine the feasibility of determining the direct relationship between RSA and SWD. The approach in this study was to utilize a TDR soil water content senor which was calibrated for mini-rhizotron tubes, allowing for the direct examination of the diurnal changes in soil water content in relation to quantified root presence. The relationship between estimated total SWD was correlated with mid-morning transpiration fluxes to validate that soil water depletion estimates were reflective of crop water uptake. The results showed a positive relationship between these variables for both genotypes (R^2=0.61 for COC 041; R^2= 0.67 for TUFRunner^{TM} '511'). These results indicate that this is a viable *in-situ* approach for quantifying SWD and RSA, allowing for mechanistic links to be made about relative differences in genotypic crop water use associated with root distribution in the soil.

The contrasting genotypic RSAs observed in this study provided suitable comparisons for examining the relationships between the structure and function of divergent root systems. The RSA of COC 041 can be generally characterized as a more fibrous root system with greater TRL of small diameter (< 3mm) when compared to TUFRunner^{TM} '511'. Additionally, the RSA of COC 041 has substantially greater TRL from 20-80 cm of soil depth than the runner market type. Despite these varying RSAs, no significant relationship was observed between TRV and SWD, when TRV was summed across all sampling depths. Similar results observing the relationship between crop water uptake and total RLD have been reported (Ratnakumar and Vadez, 2011; Zaman-Allah et al., 2011). Therefore, these results confirm that simply increasing the

total amount of root mass, even at deep soil depths, may not necessarily lead to an increased capacity for water extraction.

A plausible explanation for the lack of relationship between root system size (sum of TRV) and SWD could be due to water uptake responses to changes in soil hydrologic conditions. A general pattern in SWD distribution emerged among both genotypes in response to fluctuating soil water contents over the growing season. When soil water content was high following a soil wetting event, the greatest proportion of SWD occurred in top 30 cm of soil. As soil drying occurred, water uptake of both genotypes moved deeper in the soil profile (30-60 cm). These results suggest that a primary influence on SWD within the soil profile is strongly controlled by the soil water conditions, and the distribution of roots in relation to these dynamic SWD zones is a critical factor. The relationships between SWD and root parameters across all soil depth classifications (0-10 cm) in this study further emphasize this point. The correlation between SWD and rooting parameters was negative when VSWC was high following an irrigation event which is most likely due to the relationship between SWD and root architecture. A general RSA observation in this study was an overall increase in roots at deeper soil depths (except the 100% TUFRunner^{TM} '511' treatment). On days with high VSWC, a negative correlation between rooting parameters and SWD occurred as a result of the predominant amount of SWD occurring where fewer of roots were observed in the soil from 0-30 cm. When water uptake patterns shifted to soil depths beyond 30 cm as a response to drying, these SWD zones contained large levels of TRL, likely resulting in the positive relationship between SWD and the increased presence of roots. Overall, these results support that the most effective RSA for maximizing soil water

uptake depends on matching the root architecture to the hydrologic scenario of a particular production environment.

Studies which make the case that a more fibrous RSA with increased TRL at deep soil depths as being a critical phenotype for improving water deficit stress resilience, generally do so in the context of rainfed cropping systems where crop water demand is predominately met through water stored deep in the soil profile (Wasson et al., 2012; Lynch, 2013; Lynch and Wojciechowski, 2015). The hydrologic conditions of this study are quite the opposite, reflecting a humid peanut production climate where water deficit stress often arises quickly in surficial layers due to soils with low water retention. However, water deficit stress is generally intermittent due to crop water demand being met by in-season receipt of precipitation and irrigation application. Several studies have demonstrated that effective RSA for maximizing water uptake in supply/pulse driven environments are those with more shallow root systems which have a high density of roots in the topsoil (Schwinning and Ehlerinder, 2001; Nakhforoosh et al., 2014; Tron et al., 2015). The current study also supports these findings when comparing the surficial (0-30 cm) soil water depletion of the 100% and 60%PA irrigation treatments of TUFRunner^{TM} '511'. The 100% treatment which led to a greater density of roots systems in the surficial soils as compared to the other irrigation treatments also had a greater amount of soil water depletion in the surficial soil over the entire mid-season measurement period. These findings highlight the importance that soil water uptake distribution is highly dependent on the soil hydrological conditions, and matching RSA's with typical soil water distribution of an environment can allow for maximizing soil water acquisition.

Although the coordination of RSA to soil hydrologic conditions has the potential to maximize soil water acquisition, it is critical to note that the utilization of this water is dependent on transpiration providing the energy potential gradient controlling the ascent of xylem sap flow and root water absorption (Boyer and Kramer, 1995). Furthermore, the rate of water transport through a plant is dependent on many physiological and anatomical factors of a plant influencing hydraulic conductivity (Vadez, 2014). In general, the genotype COC 041 had lower mid-morning transpiration fluxes than TUFRunner^{TM} '511' on many of the gas exchange measurement dates in this study. The greater mid-morning transpiration flux of TUFRunner^{TM} '511' on days when the soil water content between the two genotypes was similar, is likely due to the regulation of hydraulic conductivity by COC 041 through reduction in stomatal aperture. However, there was an interaction between genotypic mid-morning transpiration rates and measurement date, a result of COC 041 having similar transpiration rates as TUFRunner^{TM} '511' on particular days. Similarities in transpiration between the genotypes typically occurred when soil water content was high following an irrigation event, and indicates that the hydraulic conductivity capability of COC 041 does have the potential to achieve similar rates of mid-morning transpiration as TUFRunner^{TM} '511'. Furthermore, this also demonstrates possible greater regulation of water flow of COC 041 caused by changes in soil water content than that exercised by TUFRunner^{TM} '511'. This is also supported by the observation of a stronger relationship between VSWC and transpiration rates with COC 041 than for TUFRunner^{TM} '511'. Variability in the inflection point at which transpiration declines due to a decline in the fraction of transpirable soil water has been reported among peanut genotypes (Devi et al., 2009). Interestingly,

despite lower mid-morning transpiration fluxes of COC 041, the rate of soil water decline was often greater than TUFRunner^{TM} ‘511’. Mid-morning transpiration fluxes were measured on the upper most mature leaf, and did not capture possible genotypic variation in transpiration rates among the lower and upper level canopy positons. For example, the erect canopy architecture of COC 041 could allow for greater light penetration and transpiration rates in the lower canopy resulting in a higher rate of soil water decline. Another possibility could be the combination of an extensive fibrous RSA of COC 041 exploring a great amount of soil volume, and greater transpiration fluxes than TUFRunner^{TM} ‘511’ following the mid-morning gas exchange measurements. Reports have documented limited-transpiration phenotypes to vary among crop genotypes which allow for increased water use efficiency and soil water conservation by reducing transpiration rates during mid-day when vapor pressure deficit is high (Sinclair et al., 2005; Devi et al., 2010; Sinclair et al., 2017).

Conclusion

There is increasing focus to assess genotypic root system architecture for identifying belowground traits which could be utilized for improving water deficit stress tolerance. However, due to the coupled nature of both above- and belowground traits it’s unlikely that assessing pertinent components of one without the other will provide fruitful information for increasing possible water acquisition and utilization under water deficit stress conditions. Additionally, environmental and management factors which influence soil hydraulic factors strongly control the suitability of genotype for a particular production system. In this study we proposed a relatively cost effective *in-situ* screening method for examining the relationship between RSA and soil water depletion distribution, which in combination with measurements of gas exchange can be used to

determine genotypic suitability for a DVS of a particular production system, or coordinated adaptive phenotyping (CAPs).

Table 4-1. Analysis of variance (ANOVA) for response variables of TRL, TRV, soil water depletion, and soil water content measured at the Plant Science Research and Education Unit (PSREU) near Citra, FL in 2016.

Effect	Total Root Length	Total Root Volume	Soil Water Depletion	Soil Water Content
IR	0.6712	0.4843	0.4805	0.8482
Geno	<0.0001	0.4075	0.0971	0.0237
IR*Geno	0.9768	0.1310	0.0640	<0.0001
Depth	0.0002	0.0080	<.0001	<0.0001
IR*Depth	0.0099	0.0001	0.7505	0.3190
Geno*Depth	0.0038	0.0303	0.0181	0.5024
IR*Geno*Depth	0.4672	0.0780	0.0038	0.8123
TM	0.0585	<0.0001	<0.0001	<0.0001
IR*TM	0.6632	0.1531	0.0044	<.0001
Geno*TM	0.4292	0.6512	0.0647	0.0349
IR*Geno*TM	0.0983	0.0182	<0.0001	0.1584
Depth*TM	0.5277	0.3693	<0.0001	<0.0001
IR*Depth*TM	0.8057	0.4350	0.6082	0.0149
Geno*Depth*TM	0.6048	0.4053	0.7068	0.9999
IR*Geno*Depth*TM	0.8795	0.6200	0.3593	0.9850

†Abbreviations: IR, irrigation treatment; GENO, genotype treatment; Depth, Soil depth increments; TM, repeated measures time within each growing season.

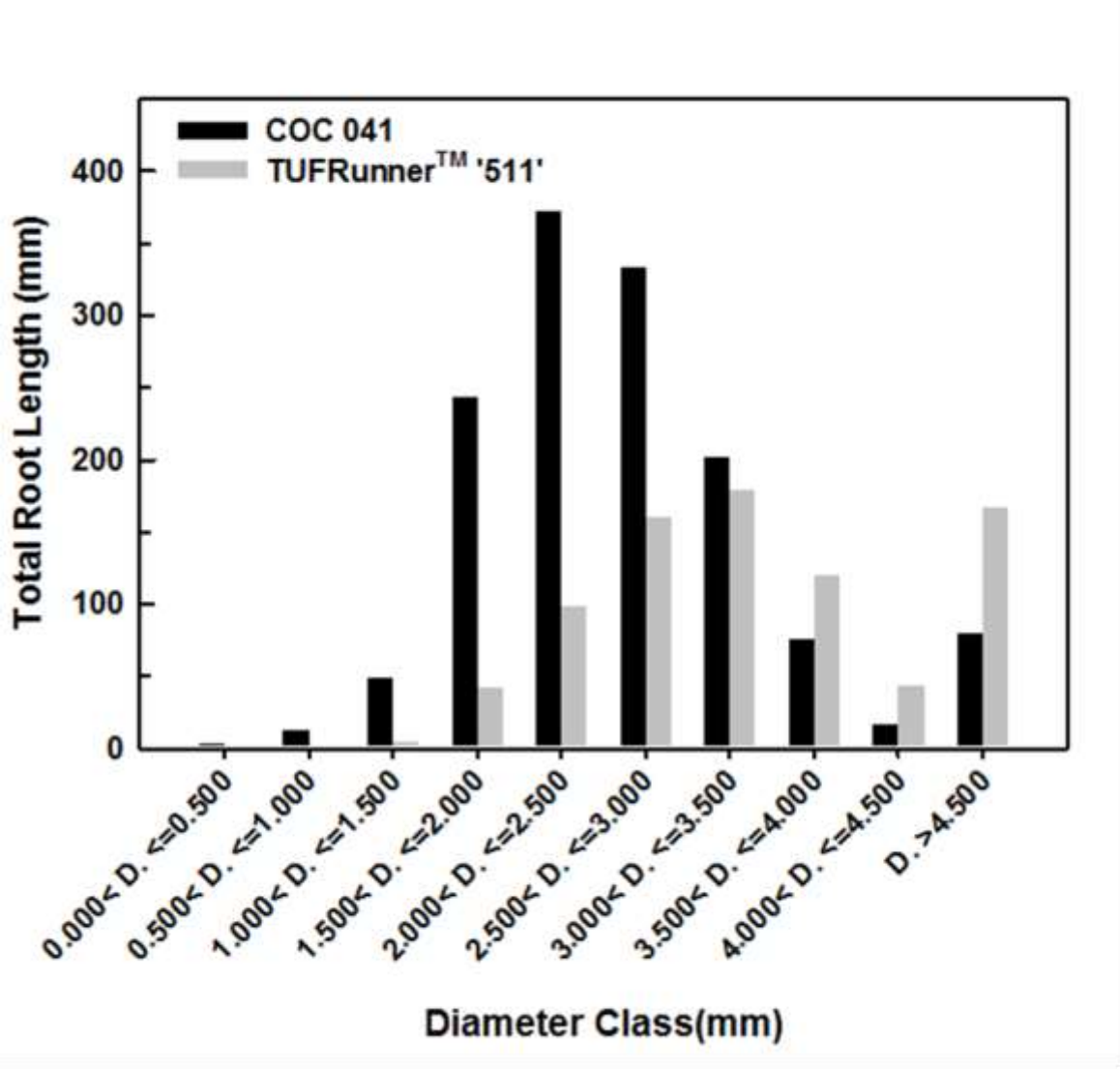

Figure 4-1. Total genotypic root length distribution for each root diameter class measures at the PSREU near Citra, FL in 2016. Total root length for each diameter class is summed to a soil depth of 80 cm.

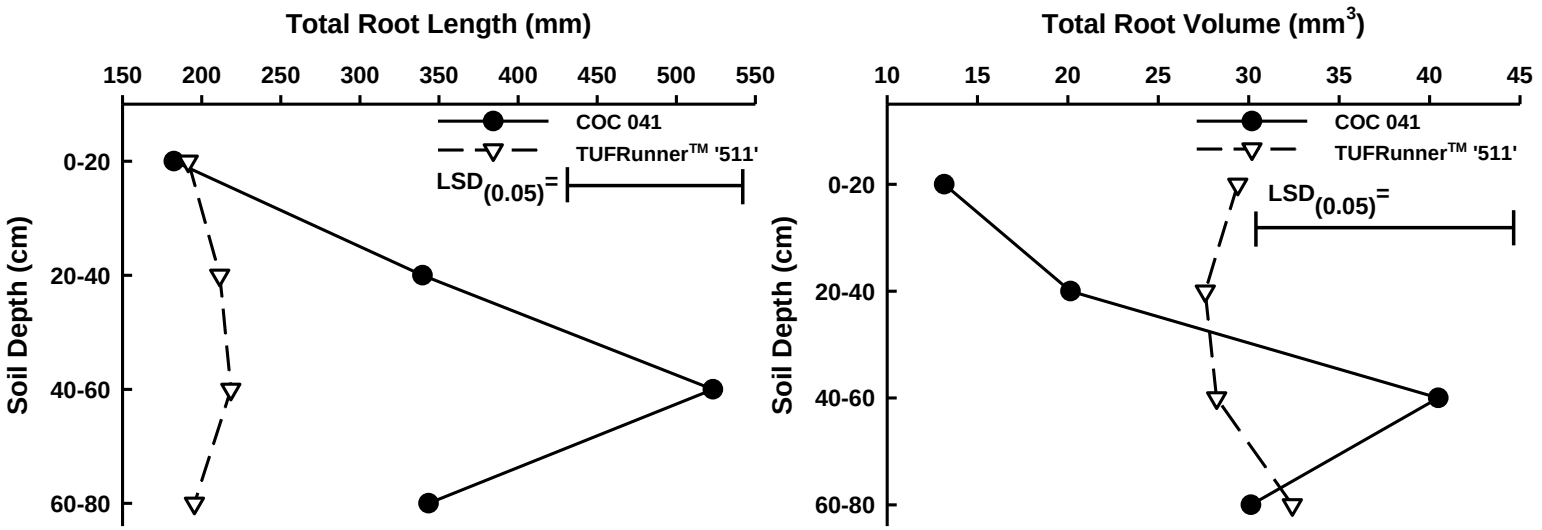

Figure 4-2. A) Average total root length of each peanut genotype by soil depth measured at the PSREU near Citra, FL in 2016. B) Average total root volume of each peanut genotype by soil depth measured at the PSREU near Citra, FL in 2016. (Abbreviations: LSD, Fischer's Protected Least Significant Difference $P < 0.05$).

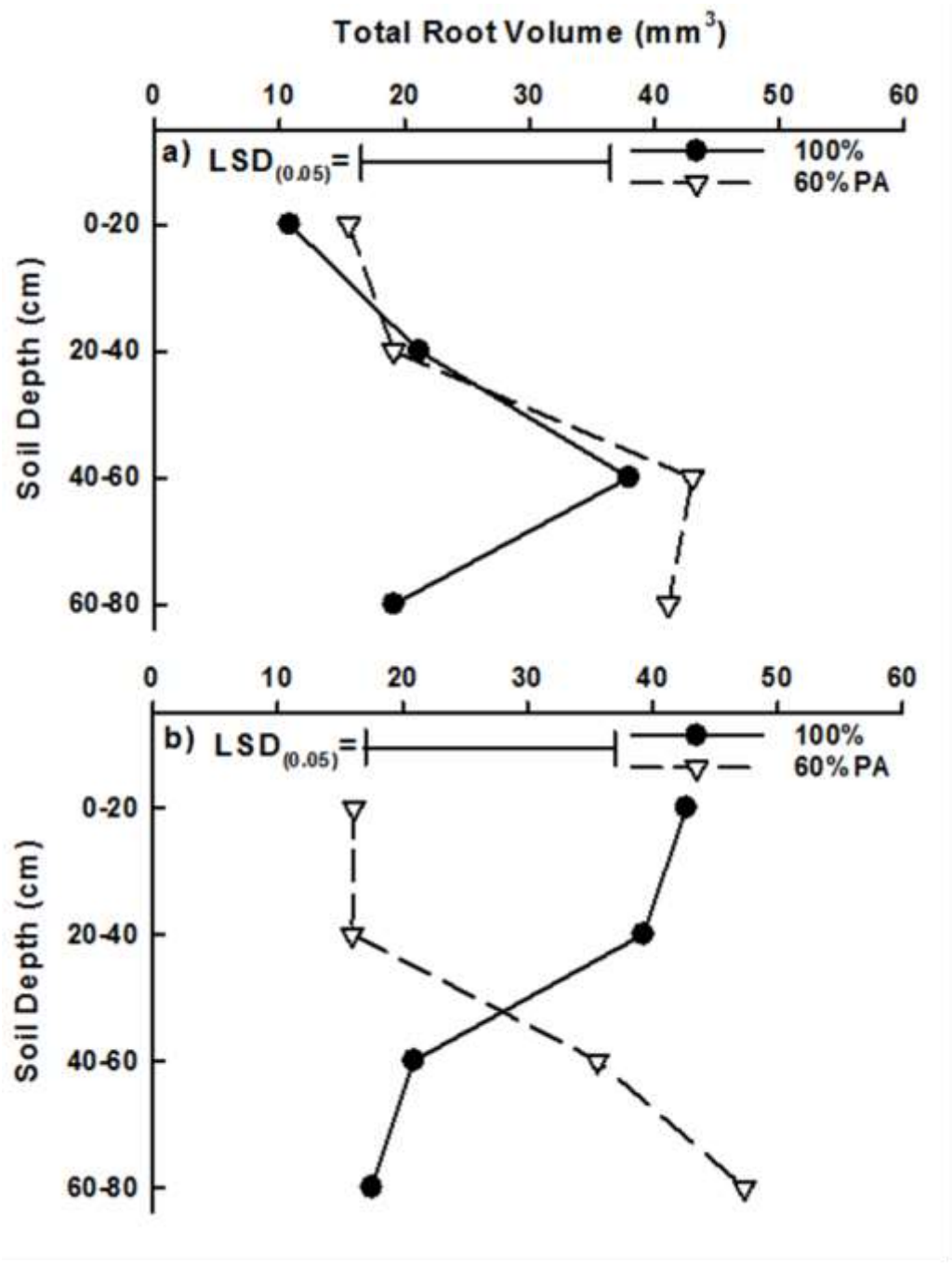

Figure 4-3. A)Total root volume architectural response of peanut genotype COC 041 to early season irrigation treatment measured at the PSREU near Citra, FL in 2016. A) Total root volume architectural response of peanut genotype TUFRunner™ '511'to early season irrigation treatment at the PSREU near Citra, FL in 2016. (Abbreviations: 100%, 1.9 cm per application; 60%PA, 1.1 cm per application until 50 DAP, followed by 1.9 cm per application for the remainder of the growing season LSD, Fischer's Protected Least Significant Difference $P < 0.05$).

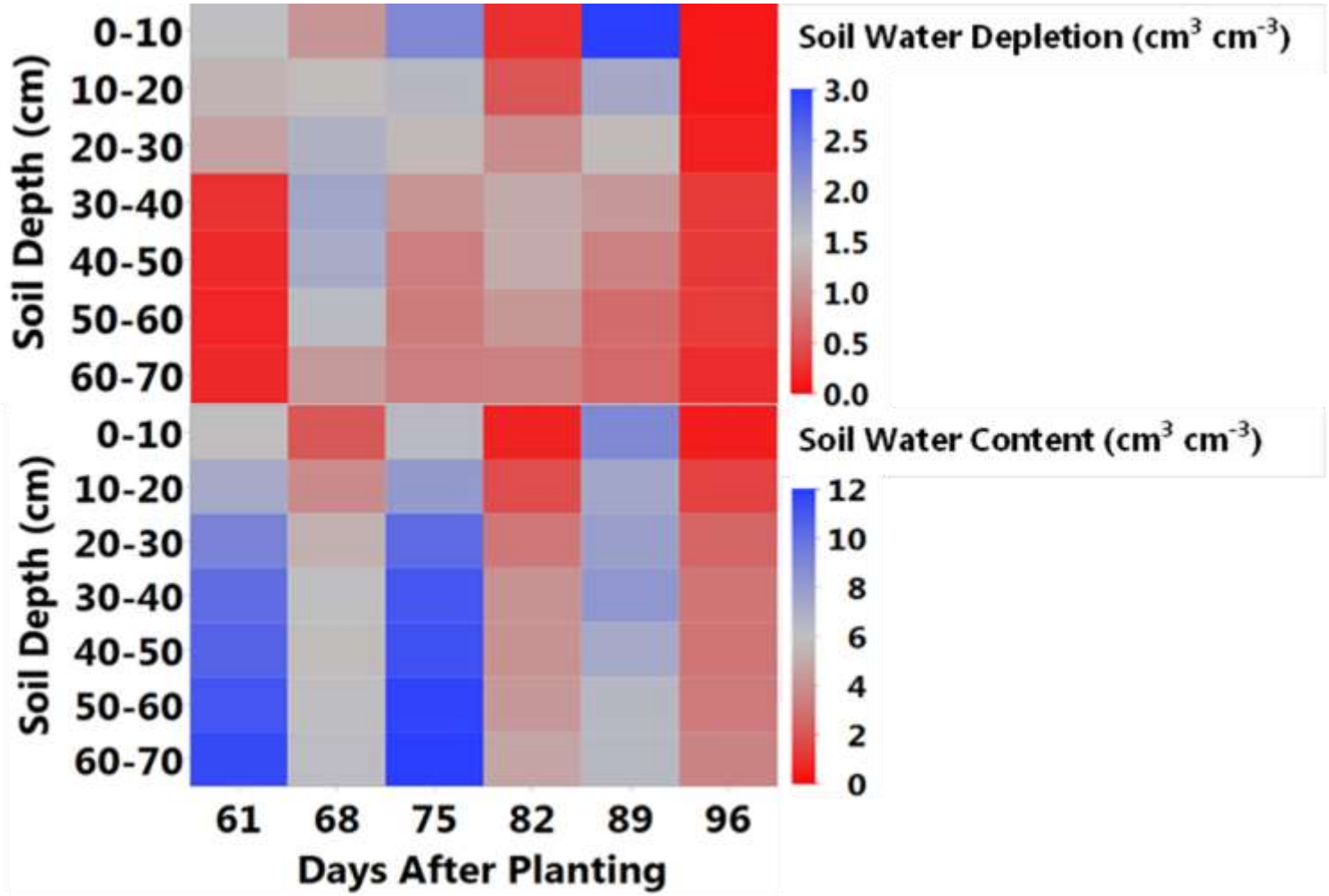

Figure 4-4. A) Soil water depletion by soil depth range for each sampling date. B) Corresponding volumetric soil water content by depth range for each sampling date. Data measured at the PSREU near Citra, FL in 2016 and represents the sampling date by depth interaction which is averaged across both peanut genotypes and irrigation treatments.

Table 4-2. Spearman correlation analysis between root parameters and soil water depletion (SWD) for each soil sampling depth on 75 DAP at the PSREU near Citra, FL in 2016. The volumetric soil water content (VSWC) on this day was high across all sampling depths and SWD was predominant in the top 30 cm of soil.

	TRL	PRA	CSRA	TRV	ARD
SWD	-0.20968	-0.21256	-0.21256	-0.18218	-0.22814
P-Value	0.0556	0.0522	0.0522	0.0972	0.0369

†Abbreviations: TRL, total root length; PA, projected root area; CSRA, cross sectional root area; TRV, total root volume; ARD, average root diameter; SWD, soil water depletion.

Table 4-3. Spearman correlation analysis between root parameters and soil water depletion (SWD) for each soil sampling depth on 82 DAP at the PSREU near Citra, FL in 2016. The volumetric soil water content (VSWC) on this day was low across all sampling depths and SWD was predominant below 30 cm of soil depth.

	TRL	PRA	CSRA	TRV	ARD
SWD	0.35132	0.32214	0.32214	0.27520	0.39756
P-Value	0.0011	0.0028	0.00281	0.0113	0.0002

†Abbreviations: TRL, total root length; PA, projected root area; CSRA, cross sectional root area; TRV, total root volume; ARD, average root diameter; SWD, soil water depletion.

Table 4-4. Analysis of variance (ANOVA) for leaf level mid-morning transpiration flux measured at the PSREU near Citra, FL in 2016.

Effect	Transpiration
IR	0.4499
GENO	<.0001
IR*GENO	0.1717
TM	<.0001
IR*TM	<.0001
GENO*TM	<.0001
IR*GENO*TM	0.0780

†Abbreviations: IR, irrigation treatment; GENO, genotype treatment; TM, repeated measures time within each growing season.

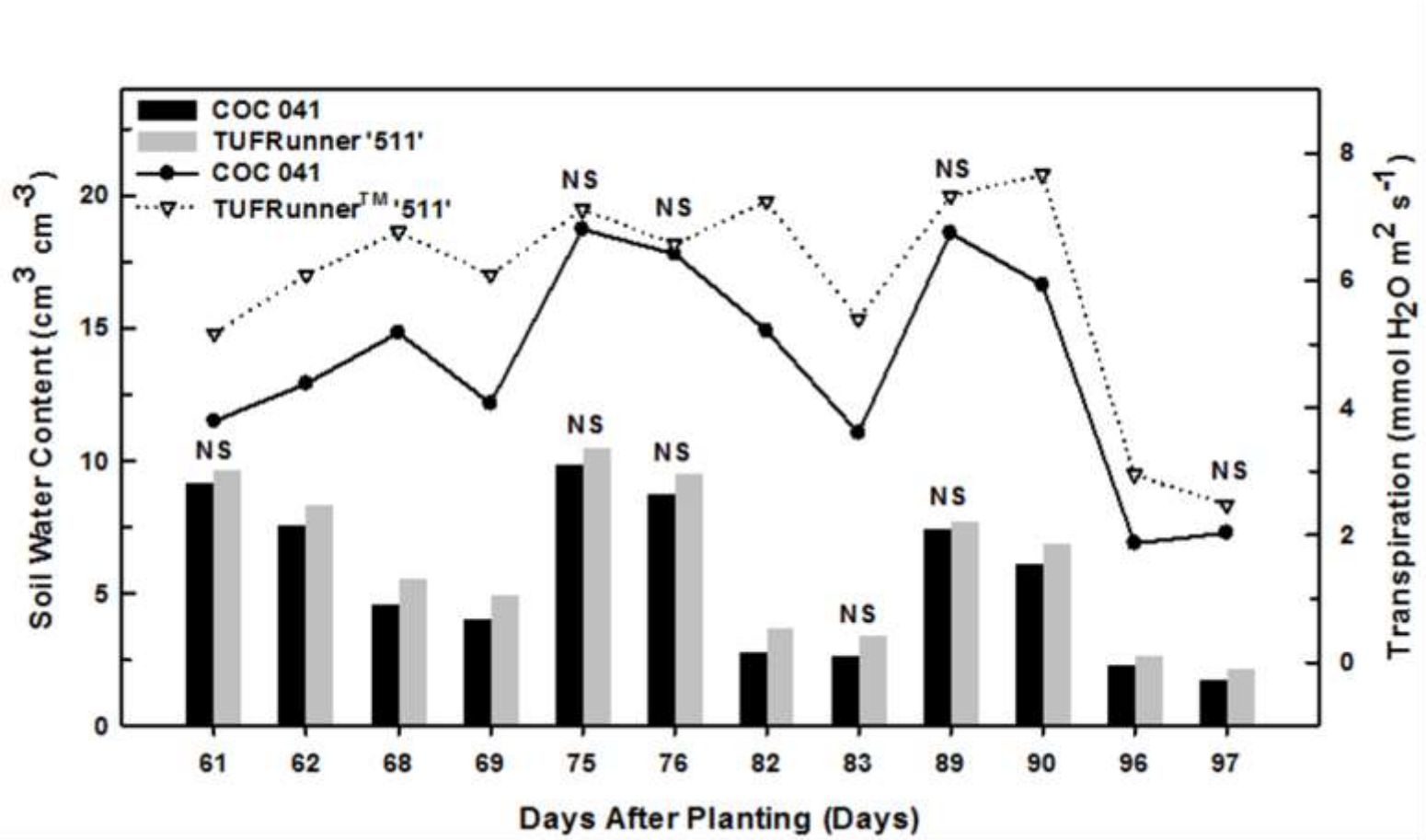

Figure 4-5. (Left y-axis) Average soil water content to a soil depth of 75 cm at each sampling date for each peanut genotype. (Right y-axis) Average leaf level mid-morning transpiration flux of peanut genotypes at each sampling date for each peanut genotype. Data was measured at the (PSREU) near Citra, FL in 2016, and is averaged across early-season irrigation treatments (Abbreviations: NS, denotes non-significance using Fischer's Protected Least Significant Difference $P < 0.05$; All other time points are significantly different using Fischer's Protected Least Significant Difference $P < 0.05$).

Table 4-5. Spearman correlation analysis between average transpiration (TR) and volumetric soil water content (VSWC) across all sampling dates for each genotype at the PSREU near Citra, FL in 2016.

	COC 041	TUFRunner™ '511'
	TR	TR
VSWC	0.4762	0.2792
P-Value	0.0033	0.0994

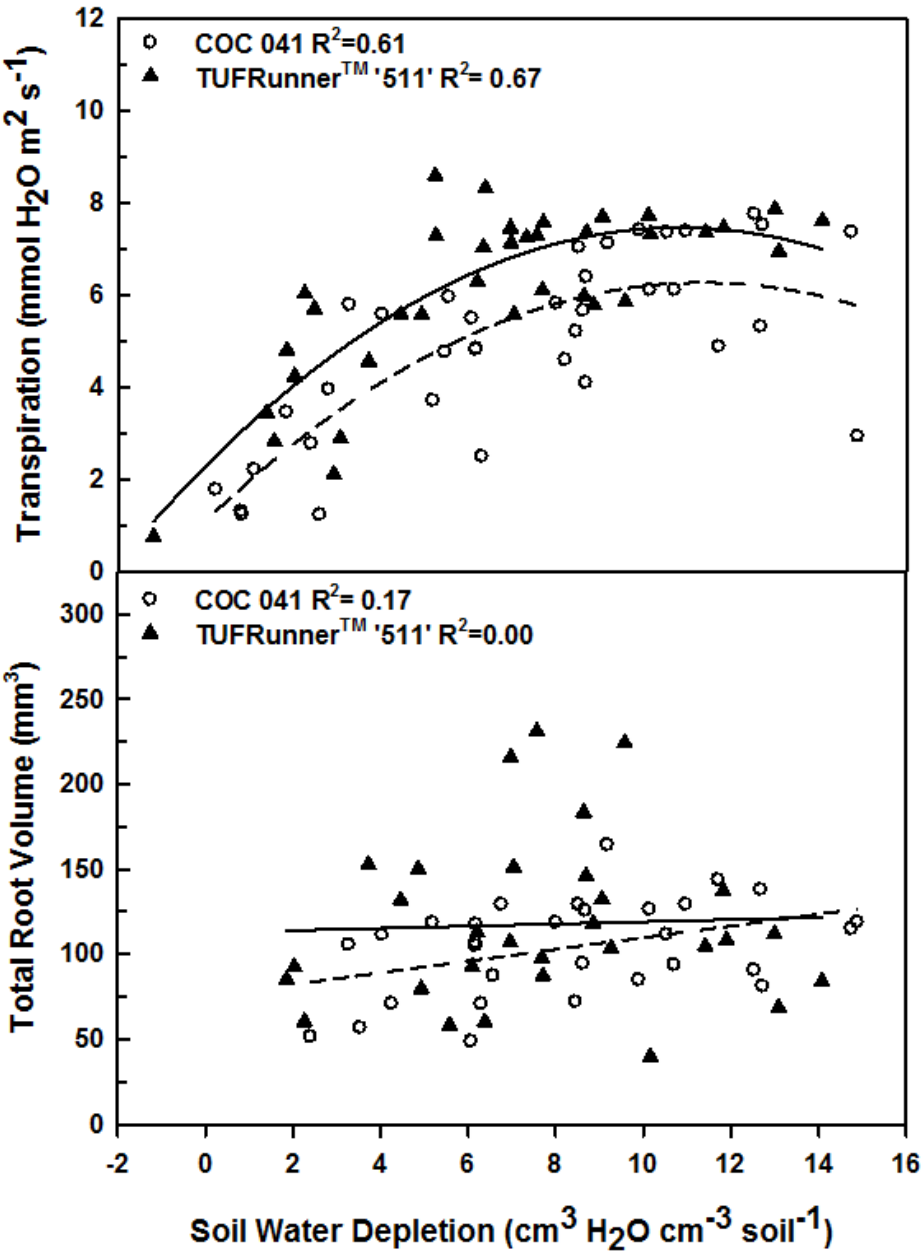

Figure 4-6. A) Relationship between leaf level transpiration and soil water depletion for each peanut genotype. Data points include all weekly sampling points from 61-96 DAP measured at the PSREU near Citra, FL in 2016. B) Relationship between total root volume and soil water depletion for each peanut genotype. Root volume is the sum of roots to a soil depth of 80 cm for each weekly sampling date.

CHAPTER 5
OPTIMIZING COTTON IRRIGATION AND NITROGEN MANAGEMENT USING A SMARTPHONE IRRIGATION SCHEDULING TOOL AND IN-SEASON NITROGEN APPLICATIONS

Introduction

Nitrogen is the most important applied nutrient in cotton production systems due to the potential of increasing lint yields by increasing N rates (Muharam et al., 2014). The possible economic gain by increasing N applications has led to many studies aimed at quantifying the N requirement per unit of lint produced, resulting in estimates ranging from 10 to 29 kg N100 kg^{-1} lint across global cotton production regions (Miley and Oosterhius, 1989; Gerik et al., 1998). Typical application rates in cotton production regions of the United States (U.S.) have reported ranges from 71-220 kg N ha^{-1} (Taylor, 1995). A major factor adding to the variability in estimates of N requirements is the overall susceptibility of N loss through NO_3^- leaching and denitrification, both which are heavily influenced by unpredictable seasonal environmental conditions and soil texture.

Using in-season N applications that split the total N applied into percentages applied at varying developmental stages is a management strategy that attempts to increase N efficiency by synchronizing soil N concentrations to plant N demand. Cotton nitrogen accumulation is non-linear with the threshold of increase reported to begin at first square, reaching peak accumulation rates at approximately early-mid flower - the developmental stage that accounts for up to 23-45% of the total N accumulation (Armstrong and Albert, 1931; Olson and Bledsoe, 1942; Bassett et al., 1970; Halevy, 1976; Oosterhuis et al., 1983; Halevy et al., 1987; Mullins and Burmester, 1991). Furthermore, N accumulation in the seeds and lint account for up to 43-60% of the total plant N, creating a large N demand sink relatively late in the growing season (Mullins

and Burmester, 2010). Geng et al. (2015) reported a yield increase of 106 kg lint ha^{-1} in one of two growing seasons when comparing a split N application (180 kg N ha^{-1} total application rate) of 40% at pre-plant and 60% at first flower compared to 100% of the N applied at pre-plant. Additionally, Yang et al. (2011) observed that when splitting N application rates of 225 kg N ha^{-1} between pre-plant and peak bloom (with a 40% application rate utilized at first bloom across all treatments), lint yields were greatest when applied at 0-40-60 % split application. Other reports have observed declines in maximal obtainable lint yields when comparing split N applications of 50% at pre-plant and first square, to yields obtained by applying 100% pre-plant N, when utilizing N rates up to 180 kg N ha^{-1} (Reiter et al., 2008). However, splitting N generally resulted in a greater amount of lint produced per kilogram of N applied translating into a greater economic return.

Soil texture has also been demonstrated to influence both the total in-season N rates and the timing of application. Some studies utilize the concept of Economically Optimal Nitrogen Rate (EONR), defined as the N rate that maximizes the value of cotton produced at a given cotton and fertilizer price. The concept of EONR is useful for evaluating the impact of N rates across soil different soil textures which have varying levels of productivity. For example, Scharf et al. (2012) reported a lower EONR for silt and sandy loams when compared to a clay soil. The EONR value was also useful for elucidating the impact of soil type on in-season N applications applied at early square growth stage. This study also evaluated different timings on in-season N applications. No yield differences were observed when N was evenly split (56 kg N ha^{-1}) between pre-plant + early square or pre-plant + early flower in sand and silty loams. However,

optimal N split for a clay soil was 1/3 pre-plant, 1/3 early square, and 1/3 early flower (Scharf et al., 2012).

Irrigation water can also be a costly management input in the approximately 40% of U.S cotton production systems which receive irrigation across the southeast and southwest regions (Vellidis et al., 2016). Total water application for producing maximum lint yields in the southwest regions of the U.S. have been reported to be 74 cm; however, the relationship between lint yield and total water applied began to predict lint yield reductions when total water applied was greater than 80 cm (Wanjura et al., 2002). However, soil texture undoubtedly influences the lint yield responses to varying irrigation regimes (Tolk and Howell, 2010; Zhou et al., 2016). Additionally, there has been research demonstrating the sensitivity of cotton lint production to timing of water stress to specific phenological stages (Hake and Grimes, 2010). Overall, these results suggest that the effectiveness of irrigation application rates on cotton growth and yield are heavily influenced by soil type, regional and local climate conditions, and phenological development, demonstrating the need to develop irrigation scheduling solutions which encompass these variables. Some current irrigation decision tools have begun to account for these variables, and are being made readily available to growers through digital irrigation scheduling tools (Migliaccio et al., 2016).

Individually, both N and irrigation management have been examined extensively in cotton; however, less research has examined these management inputs simultaneously as they occur in production systems. Pettigrew and Zeng, (2014) reported that when all N was applied prior to planting, there was no additional gain in lint yield from furrow irrigation when no N was received, although lint yield increases with

irrigation were obtained at N rates of 56 and 112 kg N ha^{-1}. Singh et al. (2010) also reported interactive effects of irrigation and in-season N management where seed cotton yield increased linearly with total N rate applications up to 200 kg N ha^{-1} with drip irrigation applied at 0.8-1.0 ET_c. In contrast, this study also reported that cotton plants treated with 0.5-0.6 ET_c achieved maximal lint yields under 160 kg N ha^{-1}, with slight yield declines at 200 kg N ha^{-1}. Contrasting observations of a lack of interaction between irrigation and nitrogen have also been reported. For example, Bronson et al. (2006) conducted a study in west Texas and found no irrigation by N interaction when irrigation was applied using a center pivot at ET replacement rates of 63-93% and N rates of a non-treated control, 164 kg N ha^{-1} applied prior to planting, and an even split application of 134 kg N ha^{-1} at pre-plant and early square.

The genetic yield potential is often never met in production systems due to a limitation of resources resulting in a failure to meet the genetic capacity for crop growth, development, and yield production (Lawlor, 2001). Management factors such as irrigation and N can have significant impacts on crop growth and production because they both have potential to influence the availability of substrates needed for photosynthesis and growth processes. For example, N provides substrate for chlorophyll production and protein synthesis which impacts both CO_2 reduction and radiation capture. Additionally, irrigation has the potential to reduce water deficit stress which can reduce CO_2 assimilation, and cell turgor reducing growth rates. Furthermore, cotton's perennial indeterminate growth habit in combination with excessive rates of N and irrigated have been reported to decrease lint yields as a result of reduced harvest index, and delayed maturity (Karam et al., 2006; Sabbe and Hodges, 2010). Therefore,

quantification of both photosynthesis and above-ground partitioning in response to irrigation and N management practices has potential to provide insight possible optimal management levels to maintain the delicate balance between having sufficient levels to avoid deficiencies, without applying excessive rates which can negatively impact yields and reduce production efficiency.

The Florida cotton production environment is characterized by soils high in sand content having excessive permeability which can result in rapid soil water depletion, and the need for supplemental irrigation to mitigate crop production losses due to water deficit stress. High intensity precipitation events can also occur during the growing season resulting in nutrient leaching and the need for in-season nutrient applications to avoid crop nutrient deficiencies. To address these issues there is a need to assess both irrigation and in-season N management for developing management strategies for growers to optimize cotton production and reduce management inputs. The overall objectives of this research were to: (i) evaluate varying levels of plant available water replacement (PAWR) estimated by the ET-based soil water balance model Cotton SmartIrrigation smartphone application (Vellidis et al., 2016) which gives site specific recommendation for irrigation scheduling; (ii) determine optimal in-season N rates when split applications are made at first square and bloom; (iii) assess the possible interactive effects of various plant available water replenish (PAWR) and in-season N rates which may influence water and N management decisions. The effectiveness of these treatments were evaluated by quantifying their influence on radiation capture by quantifying photosynthetic responses using OJIP transient fluorescence protocol, SPAD chlorophyll content, and leaf area index (LAI). Additionally, N uptake, harvest index, and

lint yield responses were measured for determining treatment impacts on nitrogen use efficiency, above-ground biomass partitioning, and yield production.

Materials and Methods

Site Characterization: Field studies were conducted during the years of 2015 and 2016 at the University of Florida's Plant Science Research and Education Unit in North Central Florida (29° 24' 38" N, 82° 10' 12" W). The soil is classified as Candler sand (Hyperthermic, uncoated Lamellic Quartzipsamments). In both years, cotton was planted into a peanut-cereal rye-cotton rotation. Soil preparation consisted of conventional tillage approximately five weeks before planting. Immediately prior to planting, the field was surface tilled using a field cultivar with S-tine sweeps. The cotton cultivar PhytoGen 333 WRF was planted at a seeding density of 10-13 seeds m^{-1} with a row spacing of 0.91 m (Dow AgroSciences, Indianapolis, IN). Plots consisted of four rows 10.6 m long. Planting was accomplished with a four-row vacuum planter (John Deere., Moline, IL). A soil test was conducted approximately six weeks prior to planting, and recommended nutrients were broadcast surface applied immediately after planting. An even split broadcast surface application was made for potassium and sulfur recommendations which consisted of ½ at planting + ½ at first square. At first flower, a tank mix of 0.6 kg B ha^{-1} and 0.22 kg a.i. ha^{-1} of pyraclostrobin (BASF, Ludwigshafen, Germany) was foliar applied. Pre-emergence weed management spray consisted of 1.1 kg a.i. ha^{-1} pendimethalin (BASF, Ludwigshafen, Germany). Post-emergence weed management spray consisted of a tank mix of 0.18 kg a.i. ha^{-1} glyphosate and 7.9 g a.i. ha^{-1} trifloxysulfuron.

Experimental Design: The experimental design consisted of a split plot arrangement in a randomized complete block design. The whole plots were irrigation

treatments of plant available water replacement (PAWR), or the percent water replenishment of the estimated root zone soil water deficit. These estimates provided by the ET-based soil water balance model Cotton SmartIrrigation smartphone application (Vellidis et al., 2016; http://smartirrigationapps.org/cotton-app/). Plant available water treatments consisted of (i) 100% of PAWR (100%); (ii) a primed acclimation (PA) treatment consisting of 50% of PAWR until first bloom and then 100% of PAWR (50%PA); (iii) 50% PAWR for the entire season (50%); (iv) a rain-fed control (RF). Irrigation was applied using a lateral move system equipped with variable rate irrigation (VRI) technology (Lindsay Corporation, Omaha, NE). Sub-plots consisted of four in-season N treatments surface broadcast applied as NH_4NO_3 at first square and flower at 0, 22, 34, and 45 kg N ha^{-1} per application. All plots received an initial N application broadcast applied as NH_4NO_3 at 22 kg N ha^{-1} immediately after planting.

Field Measurements: Leaf area index (LAI) was measured using an LAI-2200 (LI-COR, Lincoln, Nebraska) and were initiated at the first square phenological development stage (2015- 40 DAP; 2016-37 DAP), with subsequent measurements occurring bi-weekly until first fully developed boll (2015- 77 DAP; 2016- 75). A single measurement of LAI for a plot was conducted by taking one above and four equidistant readings below the canopy as a perpendicular transect between the rows with the sensor view parallel to the row and repeating this sequence with the sensor view perpendicular to the row to gain greater spatial averaging. A lens cap cover with a 45° angle was used on all measurement dates. Measurements were taken in early morning with a cast shadow over the plot area to prevent diurnal bias and underestimation of LAI values.

Leaf level measurements included SPAD chlorophyll content and OJIP chlorophyll fluorescence parameters. The SPAD measurements were initiated at first square phenological development stage with subsequent measurements occurring bi-weekly until approximately first fully developed boll (Konica Minolta, Tokyo, Japan). These measurements occurred on the most newly formed mature leaf. Ten plants were randomly selected per plot and averaged. The OJIP measurements were initiated at first developed boll in both years, dark acclimated transient chlorophyll fluorescence (OJIP) was measured and recorded weekly until boll opening (2015-112 DAP; 2016-110 DAP) using a pulse-modulated fluorometer (Optisciences, Hudson, NH). Two plants per plot were measured using the most newly formed mature leaf and averaged. Measurements were recorded in the evening (18:00-20:30), and dark acclimation clips were placed on leaves for approximately 60 minutes. Saturated light intensity was set for 3500 µmols with a time duration of 3 seconds. Transient chlorophyll fluorescence parameters were calculated in as described by Stirbet and Govindjee, (2011). Parameters calculated were (i) quantum efficiencies/probabilities of ψ_{ET2o}, $\psi_{RE1o,}$ δ_{RE1o}; (ii) Specific energy fluxes per reaction center of J^{abs}/RC, $J_o{}^{DI}/RC$, $J_o{}^{TR}/RC$, $J_o{}^{RE1}/RC$, RC/J^{ABS}; and (iii) Phenomenological energy fluxes per cross section of J^{abs}/CS_o, $J_o{}^{DI}/CS$, $J_o{}^{TR}/CS$, $J_o{}^{RE1}/CS$, RC/CS (Table 5.1) (Figure 5.1).

When all plots had approximately 30% of their bolls open by visual assessment, 0.75 m of row in each plot was harvested for determining N uptake by cutting the stems at the soil surface and fresh above-ground biomass per unit area was determined. Whole plants were ground using a wood chipper and approximately 500 grams of fresh weight was collected as a sub-sample. This sub sample was dried at 70° C for seven

days and weighed for determining tissue water content which was used to calculate the dry above ground biomass per unit area. The dry sub-sample was further ground through a Wiley Mill and plant material that passed through a 1mm sieve was collected. This plant tissue was analyzed for total N (Waters Agricultural Laboratories, Inc., Camilla, GA) using the combustion method. Total percent N content was multiplied by the dry above-ground biomass per area to determine total N uptake per area. Decision to terminate cotton was done using the four nodes above the cracked boll method, and a harvest aid of 0.56 kg a.i. ha^{-1} paraquat dichloride was sprayed for defoliation. The two center rows of each plot were machine harvested using a spindle cotton picker (John Deere, Moline, IL) for lint yield on 139 and 162 DAP in 2015 and 2016, respectively. The spindle picker was modified so cotton lint for each plot could be collected in mesh bags. Cotton lint collected was hand weighed for each plot and a 250 gram sub-sample was collected and the seed removed from the lint using a table top cotton gin (manufacturer, location) to determine lint yield alone (gin turnout). The determined lint weight per area and above-ground biomass per area harvested at 30% open bolls was used for determining harvest index. Harvest index was calculated as the ratio of lint weight per area to total above-ground biomass per area.

Statistical Analysis: Statistical analysis was performed using SAS v. 9.4 statistical software (SAS Institute, 2013). PROC GLIMMIX was used to compute analysis of variance (ANOVA). Random effects of rep nested within year, and rep crossed with irrigation nested within year were included in the model. Repeated measures ANOVA was performed on measurements that were repeated over the growing season (SPAD, LAI, and chlorophyll fluorescence). Autoregressive covariance

matrix structure was specified for repeated measure ANOVA's. Normality and homogeneity were visually assessed by graphing the residual distribution, scatter plot of residuals, and Q-Q plot of residuals. No data was pooled over the two site years due to the strong main effect of year. Data was pooled (when specifically) over other factors when appropriate as indicated by the F-test results. Multiple comparisons significance was determined using Fisher's Protected Least Significant Difference (LSD) at the $P < 0.05$ probability level.

Results

Yearly precipitation and irrigation: Contrasting amounts of precipitation were received during the two growing seasons of 2015 and 2016; total cumulative rainfall received in 2015 and 2016 was 837 and 647 mm, respectively (Figure 5.2). The varying amounts of precipitation impacted the total number of irrigation events scheduled within the two growing seasons. In 2015, a total amount of three irrigation applications were applied within the range of 41-63 days after planting (Table 5.2) for a total of 59 mm. Two of these irrigation applications occurred prior to first bloom. The third and final irrigation treatment was applied seven days after first bloom. In 2016, nine irrigation applications were applied ranging from 31-84 DAP for a total of 205 mm. Three of these applications occurred prior to first bloom, four in between first bloom and developed boll, and two in early boll development.

Leaf area index and SPAD chlorophyll content: Differences in total water received in each year likely led to the differences in LAI between years (Table 5.3). What was similar between years was an observed trend where LAI continued to increase up until six weeks after first square (82 and 79 DAP in 2015 and 2016, respectively) to values of 4.7 and 2.9 (averaged across treatments) in 2015 and 2016,

respectively (Figure 5.3 & 5.4). Following six weeks after first square, a decrease in LAI occurred in both years likely due to leaf senescence, to reductions of 32 and 19% in 2015 and 2016, respectively. Year also interacted with the irrigation and N treatments: in 2015, the 100% irrigation treatment had a greater LAI than all other irrigation treatments at two weeks after first square (Figure 5.3). Additionally, the 50%PA treatment in 2016 resulted in a greater LAI at the six and eight weeks after first square measurements when compared to the other irrigation treatments (Figure 5.4). For N treatments in both years, all treatments which received in-season N (22, 34, and 45 kg N ha^{-1}) maintained a greater LAI than the non-treated control following first bloom; further, these in-season N treatments maintained similar LAI across the entire season in 2015. Results were similar in 2016, with the exception of increased LAI when comparing the 34 kg N ha^{-1} to the 22 kg N ha^{-1} treatment at six weeks following first square.

Differences in water treatments were not evident in 2015 for SPAD chlorophyll; whereas in 2016 the RF and 50% irrigation treatments maintain greater SPAD chlorophyll content later in the growing season when compared to the 100% and 50%PA treatments (Figure 5.3). Side-dress N rates did impact SPAD chlorophyll content in 2015 which resulted in the 22 kg N ha^{-1} treatment having lower chlorophyll content than the 45 kg N ha^{-1} at six and eight weeks after first square. The 45 kg N ha^{-1} treatment had similar SPAD chlorophyll content to the 34 kg N ha^{-1} treatment. In 2016, the SPAD chlorophyll content was similar among the side-dress N treatments, except for the last measurement date at 8 weeks after first square (Figure 5.4). Overall when averaged across all treatments, there was a lower SPAD chlorophyll content during the

2015 growing season with a marked decline after first square in this year, whereas SPAD chlorophyll content was relatively stable over the measurement period in 2016.

Transient Chlorophyll Fluorescence (OJIP): Side-dress N affected the energy fluxes of PSII photochemistry when measured during boll development in the 2015 growing season (Table 5.5). Phenomenological energy fluxes of $J^{ABS/CS}$, $J_o^{DI/CS}$, and $J_o^{TR/CS}$ were increased when comparing the greatest N rate (45 kg ha^{-1}) to the treatment which received no in-season N. The other two reduced N rates (22 and 34 kg N ha^{-1}) were similar to the 45 kg N ha^{-1} treatment. These same energy fluxes expressed on a reaction center (RC) basis had a reverse trend where the non-treated control had increased values when comparing to the greatest side-dress N rate at 45 kg ha^{-1}. No differences were observed in the specific energy fluxes among the N treatments which received in-season N. The amount of reaction centers per cross sectional area (RC/CSm) increased with increasing N rates. Increased N rates also had a similar trend of increased electron transport flux until PSI acceptors on both a reaction center $J_o^{RE1/RC}$ and cross sectional basis $J_o^{RE1/CS}$. In 2016, there was little influence of N treatment on the phenomenological or specific energy fluxes. No differential influences of N treatments were also observed in the amount of RC/CSm or $J_o^{RE1/CS}$.

Irrigation treatments had little influence on the efficiency of electron transport during the 2015 growing season, while in 2016, there was in interaction between irrigation and date (Figure 5.5). These interactions between irrigation and date may have been stronger for 2016 due to the relatively drier conditions experienced in this year requiring more frequent irrigations (Figure 5.2), particularly during boll development. At first boll and one week after first boll, φ_{RE1o} was greater when

comparing all treatments which received irrigation to the RF treatment, but by the third week into boll development these relationships started to converge with the 100% irrigation treatment having greater δ_{RE1o} when compared to the RF treatment (Figure 5.5). Within the electron transport chain, the transport efficiency from Q_a to Q_b (φ_{ET2o}) was elevated in the 60%PA treatment at the two and three week sampling compared to the RF. Examining the φ_{RE1o} relationships among irrigation treatments show a similar relationship to $\varphi_{RE1o,}$ indicating that the mechanisms which drive electron transport efficiency from PSII to PSI are behaving differently depending their plant water status.

Nitrogen Uptake: There was a direct effect of N application rate on the total N uptake in both years, with no interaction between N and irrigation (Table 5.5) A reduced amount of N uptake was observed in the treatment that only received the base application at planting (22 kg N ha^{-1}) compared to all the other treatments in both years (Figure 5.6). When comparing treatments that had received in-season N, the patterns were different among years. For 2015, no differences in N uptake existed; while in 2016, the two mid-level treatments (22 and 34 kg N ha^{-1}) differed. When averaged across all treatments there was an increase in N uptake of 56 kg N ha^{-1} when comparing 2016 to 2015.

Lint Characteristics and Harvest Index: Both N and irrigation had an interactive effect with year on lint yield (Table 5.5). In the relatively wet year of 2015, no lint yield differences were observed among irrigation treatments (Figure 5.7). However, during the relatively dry year of 2016, the 100% and 60%PA irrigation treatments had the greatest yields. For N treatments, 2015 showed differences in yield only between the 22 and 112 kg N ha^{-1} treatments, with the highest yield measured in the latter

treatment. Surprisingly, the 112 kg N ha^{-1} treatment in 2016 had significantly lower yield than the other three application rates, even though this reduced yield was not reflected in the tissue N content. Influences of these yield patterns were evident in the HI as expected, since HI is calculated using yield. The impacts of irrigation were only evident in 2016, with an overall lower HI for the RF treatment (Figure 5.7). Surprisingly, the pattern of HI in 2015 was opposite to that of yield: while the lowest N treatment had the lowest yield in that year, HI was greatest for this treatment. This indicates that the relative allocation to aboveground biomass in the 22 kg N ha^{-1} treatment would have also significantly reduced total biomass, resulting in an elevated HI even though yield was low. For 2016, the yield and HI patterns were similar, with the highest and lowest HI values recorded for the 22 and 112 kg N ha^{-1} treatments, respectively (Figure 5.6).

Discussion

The contrasting environmental conditions among the two years of this study allowed for an opportunity to examine contrasting climatic conditions which can make cotton N management challenging on the marginal sandy soils of north central Florida. The large amounts and intensity of precipitation events in 2015 occurring subsequent to both in-season N applications were conducive for considerable NO_3^- leaching. This was evident from the continual decrease in SPAD chlorophyll content in all N treatments following the last N application at first bloom (54 DAP). However, the greater amount of N uptake in treatments which received in-season N prior to the period of intense rainfall at 63 DAP likely contributed to greater LAI.

Late season plant N status had implications on photosynthetic performance (as measured by OJIP) in 2015. This is a critical period of boll development (77-112 DAP) when carbohydrate demand to support the formation and maturation of bolls is high.

Under N limiting conditions, fewer PSII RC/CS were formed which was likely due to the high N demand required for synthesizing individual PSII reaction centers proteins. To compensate for fewer PSII reactions centers, the data indicated that larger antenna complexes were likely associated with each reaction center, resulting in a greater amount of trapping per reaction center (J_o^{TR}/RC). But the data also indicate that this compensation mechanism of larger antennae complexes was unsuccessful at increasing electron transport to the acceptors of PSI. This is shown by the greater amount of $J_O^{RE1/}$RC when comparing the 45 to the 22 and 0 kg N ha^{-1} treatments, indicating that electron transport may be limited by intermediate steps between PSII and PSI in the 22 and 0 kg N ha^{-1} treatments. This may be due to N deficient plants having less Rubisco production which would result in decreased carboxylation, ATP synthase and NAD(P)H, causing an overall down regulation of linear electron transport flux to PSI acceptors (Theobald et al., 1998).

In 2015, two early season irrigation applications made prior to first bloom, followed by one irrigation application after first bloom, resulted in increased LAI in the 100% irrigation treatment until peak LAI's were reached. However, the abundance of late season precipitation resulted in no late season irrigation applications. This resulted in no differences in photosynthetic performance among irrigation treatments during boll development. Despite no differences in photosynthetic performance, the 100% treatment also had increased lint yield when comparing the 50% and RF irrigation treatments, however the 50%PA yielded similarly to the 100%, despite a lower LAI. These results indicate that the yield decline when comparing the 50% treatment to the 100% treatment can be attributed to the third and final irrigation treatment which

occurred at approximately peak bloom. Peak flower has been documented to be a sensitive time for water stress due to the greatest number of young bolls being present on the plant, which are most sensitive to water deficit induced abscission (Grimes et al., 1970; Hake and Grimes, 2010). It also suggests that increased canopy growth in the 100% irrigation treatment did result in additional lint yield when compared to the 50%PA, indicating that the yield decline of the 50% and RF treatments was due to other factors than canopy growth, possibly a decreased number of bolls formed due to water stress disrupting cotton flower pollination. This would be consistent with studies showing that increasing water stress severity increases square/boll shedding and total number of bolls, thus reducing total lint yield (Cetin and Bilgel, 2002; Singh et al., 2010; Lockhande and Reddy, 2014) Nonetheless, these results indicate that a reduction of up to 50% PAW replenishment can occur until first bloom in this sandy soil type without a decline in yield.

The dry season of 2016 had little response in LAI with increasing in-season N. The absolute values of LAI were much lower when comparing 2015 to 2016 (2015-3.0; 2016-2.0). This was likely due to a reduction of carbon assimilation from water deficit stress as a result low precipitation during mid-season cotton development. Late season N deficiency was not evident from SPAD chlorophyll readings, and approximately 56 kg N ha^{-1} more uptake when comparing average values in 2016 to 2015. In 2016, increasing N rates resulted in decreased HI values, most starkly evident in the 45kg N ha^{-1} treatment that had a decreased lint yield of 56 kg lint ha^{-1}. A combination of increased plant N levels and late season precipitation may have led to excess late season canopy growth and a delay in lint maturation in 2016. Regardless of the relative

wet or dry conditions within a growing season these results show no additional yield gains were made when applying more than 22 kg N ha^{-1} in uniform split applications made at first square and bloom. Therefore, in years where precipitation and leaching potential is high on these sandy soils, it is likely that additional gains in lint yield from N management may only be made by increasing the number of N applications, not increasing the application rate.

The very dry year of 2016 further supports the conclusion that a 50% reduction in PAW until first bloom can be implemented without a reduction in yield. Three applications were made prior to first bloom, resulting in canopy growth differences among the treatments which received irrigation. Interestingly, the 50%PA irrigation treatments reached the greatest maximal LAI, despite receiving similar amounts of irrigation as the 100% irrigation treatment following first bloom. However, increased canopy growth did not correspond to increase or decreases in lint yield between the 50%PA and 100% irrigation treatments. Whereas, the 50% and 100% had similar seasonal LAI values, but a yield decrease occurred with the 50% irrigation treatment. Additionally, δ_{RE1o} were similar when comparing the 100%, 50%PA, and 50% irrigation treatments during boll development, indicating that the photosynthetic performance at this time was similar. These results show that the overall yield reduction in the 50% treatment may have been due to water stress impacts on reproductive development prior to the first formed boll, and even greater lint yield reductions occurred in the rainfed treatment where a reduction in δ_{RE1o} was observed during the first two weeks of boll development. In both years, the research supports that a 50% reduction in PAWR can occur up until first bloom without any detriment to yield. However, when a 50%

reduction in irrigation was extended past first bloom lint yield reductions did occur when compared to 100%PAWR, highlighting that water savings management should be targeted prior to first bloom.

Conclusion

The contrasting climatic condition between years allowed for a strong assessment of both N and irrigation management. Under both years, lint yield maintenance or gains occurred with making two split N applications at first square and bloom of 22 kg N ha^{-1}, a 45 kg N ha^{-1} reduction in comparison to the traditional total N recommendation of 112 kg N ha^{-1}. Yield maintenance also occurred when reducing the PAWR up until first bloom. These management strategies offer ways to cut input cost which are of economic importance when growing cotton in marginal production environments which often have an overall lower yield potential.

An interesting research finding of this research was the lack of interaction between N and irrigation management on canopy development and yield. Particularly, in the dry year of 2016 this lack of interaction supports that the soil water content conditions did not limit N availability and uptake, and that increased canopy growth due to N application did not exacerbate water stress. A possible explanation for these results could be differential root: shoot growth responses to both irrigation and N management. Further investigation into the interactive phenomenon of root: shoot growth responses, and synergisms of both irrigation and N management under field conditions are needed.

Table 5-1. Transient chlorophyll fluorescence (OJIP) parameter definitions as defined by Stirbet and Govindjee, (2011).

OJIP Parameter	Quantum Efficiencies
ψ_{RE1o}	efficiency that a PSII trapped electron is transferred until PSI acceptors
ψ_{ET2o}	efficiency that a PSII trapped electron is transferred from Q_a to Q_b
δ_{RE1o}	efficiency that a PSII trapped electron is transferred from Q_b to PSI acceptors
	Specific energy fluxes per reaction center
J^{abs}/RC	average absorbed photon flux per PSII reaction center
J_o^{DI}/RC	dissipated energy flux in processes other than trapping per PSII reaction center
J_o^{TR}/RC	maximum trapped exciton flux per PSII reaction center
J_o^{RE1}/RC	electron transport flux until PSI acceptors per PSII reaction center
	Specific energy fluxes per cross section
J^{abs}/CSo	average absorbed photon flux per cross section
J_o^{DI}/CS	dissipated energy flux in processes other than trapping per cross section
J_o^{TR}/CS	maximum trapped exciton flux per cross section
J_o^{RE1}/CS	electron transport flux until PSI acceptors per cross section
RC/CS	number of active PSII reaction centers per cross section

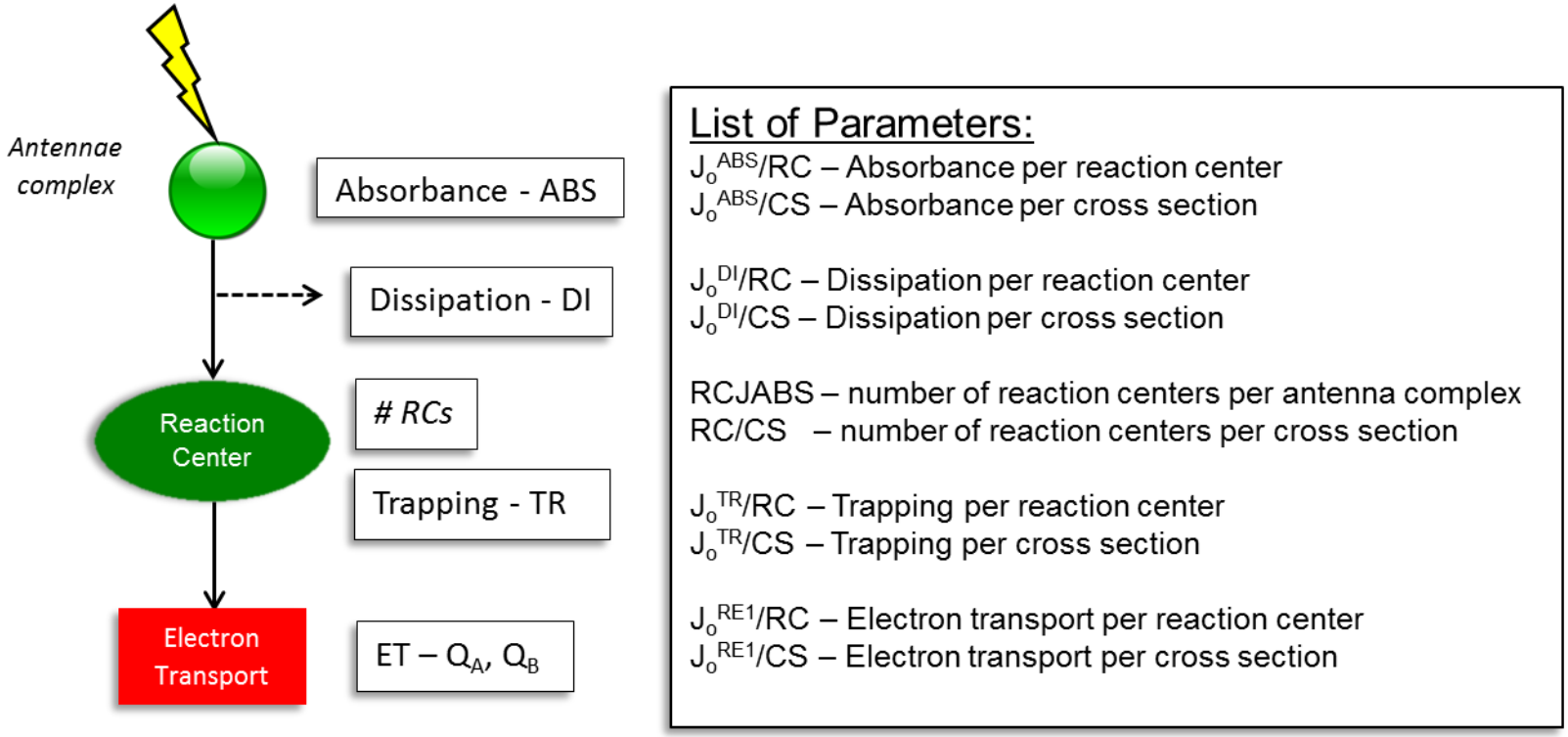

Figure 5-1. Depiction corresponding transient chlorophyll fluorescence (OJIP) parameters to the location within the light reactions of photosynthesis. Physical energy of light is absorbed by the antenna chlorophyll complex which is associated with a reaction center. Part of the absorbed energy is dissipated as heat and fluorescence (dashed line). Energy which is channeled to the reaction center is 'trapped' and converted to redox energy by electron transport chain.

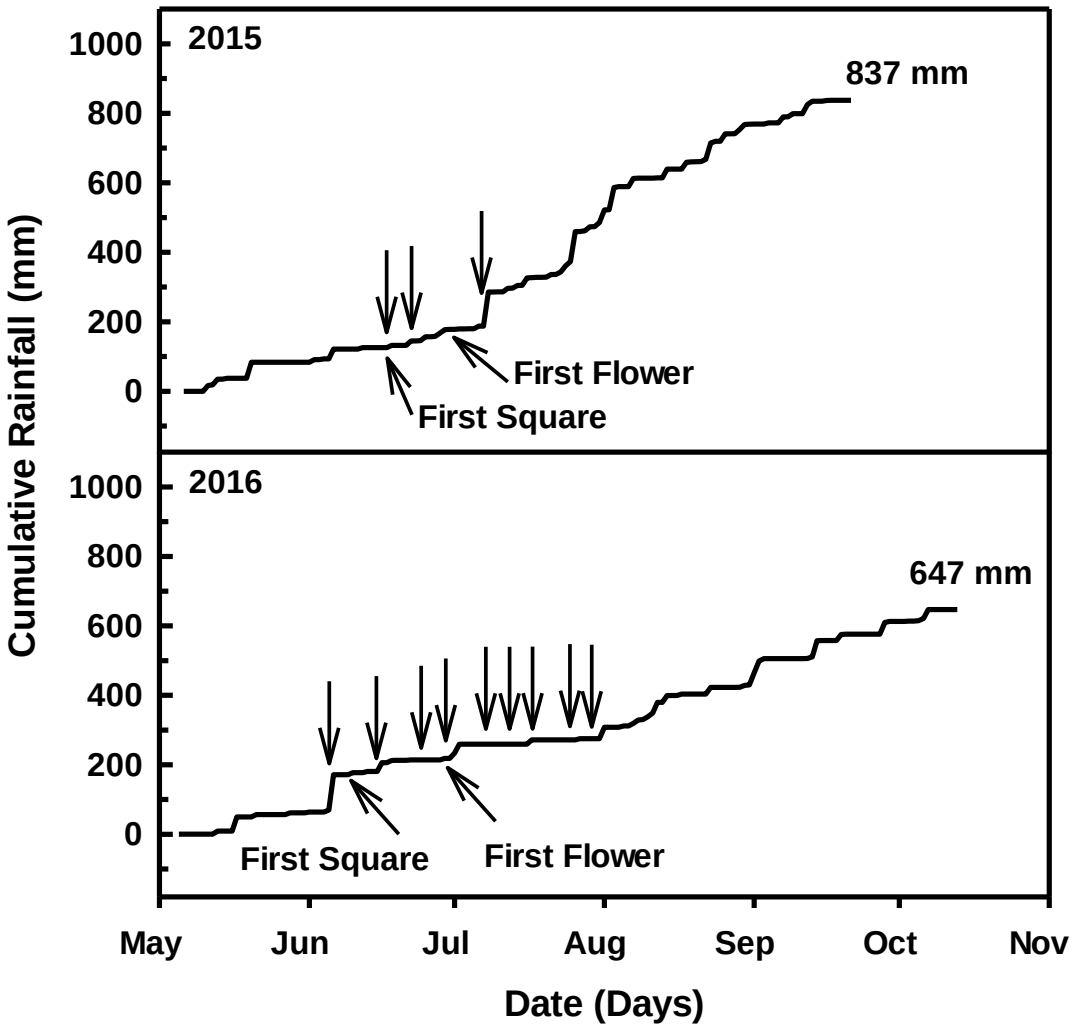

Figure 5-2. Cumulative precipitation from 1 May through 1 November for 2015 and 2016 at the Plant Science Research and Education Unit (PSREU) near Citra, FL. Cumulative rainfall axis starts at planting for both years. Vertical arrows indicate when irrigation was applied.

Table 5-2. Irrigation and total water received during the 2015 and 2016 growing season for each irrigation treatment at the PSREU near Citra, FL. Total water received is the sum of irrigation and precipitation.

	Irrigation Water Applied		Total Water Received	
	2015	2016	2015	2016
Irrigation Treatment	mm			
100%	59	204	896	851
50%PA	46	179	883	826
50%	30	102	867	749
Rainfed	0	0	837	647

† Abbreviations: 100%, 100% PAWR per irrigation application; 50%PA, 50% PAWR per irrigation application until first flower followed by 100% PAWR per irrigation application; 50%, 50% PAWR per irrigation application for entire growing season; RF, rainfed control.

Table 5-3. Analysis of variance (ANOVA) for SPAD chlorophyll content and leaf area index (LAI) variables at the PSREU near Citra, FL.

Effect	SPAD	LAI
year	<0.0001	<0.0001
IR	<0.0001	<0.0001
year*IR	0.0288	00.0004
N	<0.0001	<0.0001
year*N	0.0127	0.1736
IR*N	0.8453	0.3829
year*IR*N	0.0917	0.8467
TM	<0.0001	<0.0001
year*TM	<.0001	<0.0001
IR*TM	0.0028	0.0005
year*IR*TM	<0.0001	<0.0001
N*TM	0.0006	<0.0001
year*N*TM	0.8589	0.3582
IR*N*TM	0.9908	0.9746
year*IR*N*TM	0.9621	0.9912

†Abbreviations: year, growing seasons of 2015 and 2016 at the PSREU; IR, irrigation treatment; N , in-season N treatment; TM, time repeated measures within each growing season.

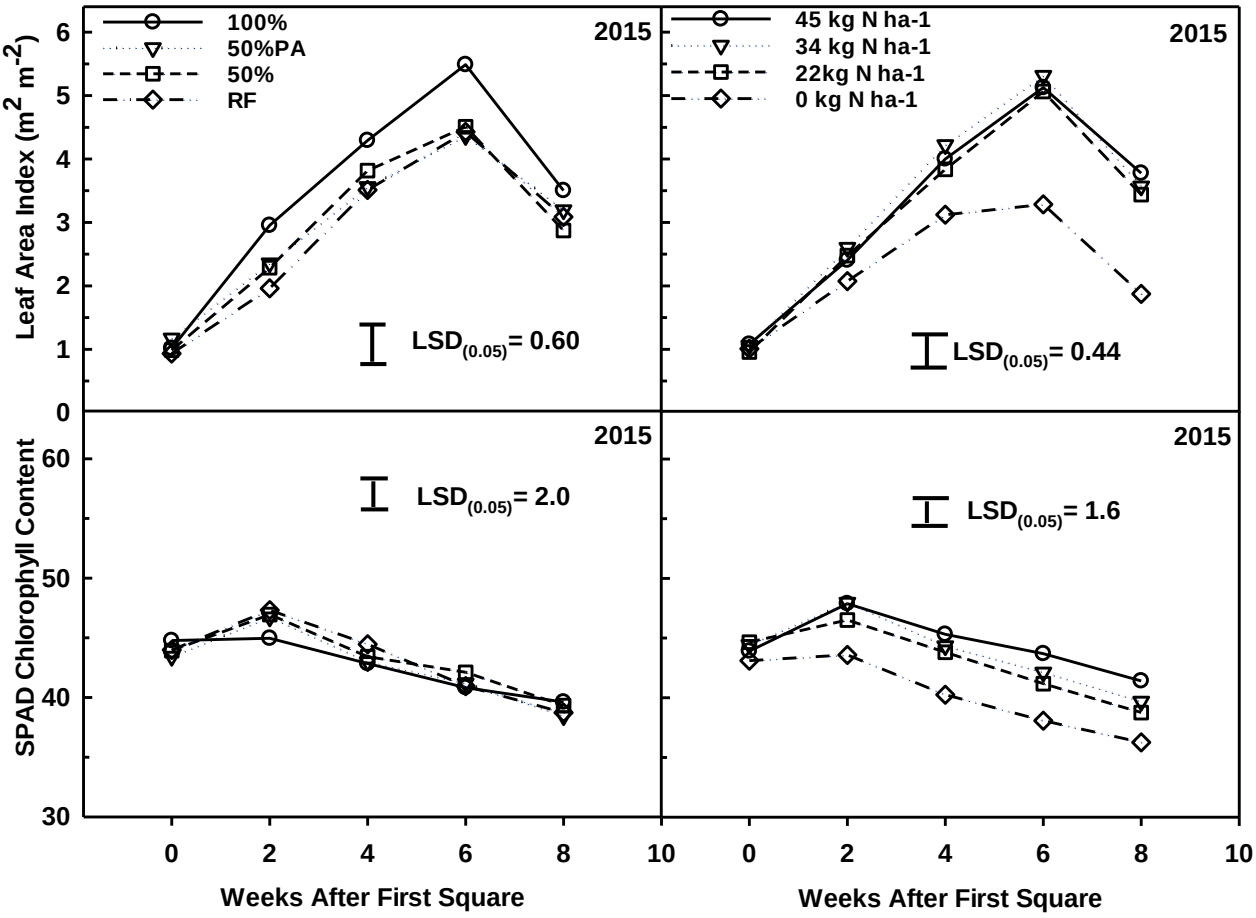

Figure 5-3. Top) Average leaf area index (LAI) starting at first square and measured until 8 weeks after first square for the irrigation and N treatments in 2015 at PSREU. Bottom) Average SPAD chlorophyll content starting at first square and measured until 8 weeks after first square for the irrigation and N treatments in 2015 at PSREU (Abbreviations: 100%, 100% PAWR per irrigation application; 50%PA, 50% PAWR per irrigation application until first flower followed by 100% PAWR per irrigation application; 50%, 50% PAWR per irrigation application for entire growing season; RF, rainfed control; Nitrogen rates are uniform in-season N rates applied at first square and bloom; LSD, Fischer's Protected Least Significant Difference at $P < 0.05$).

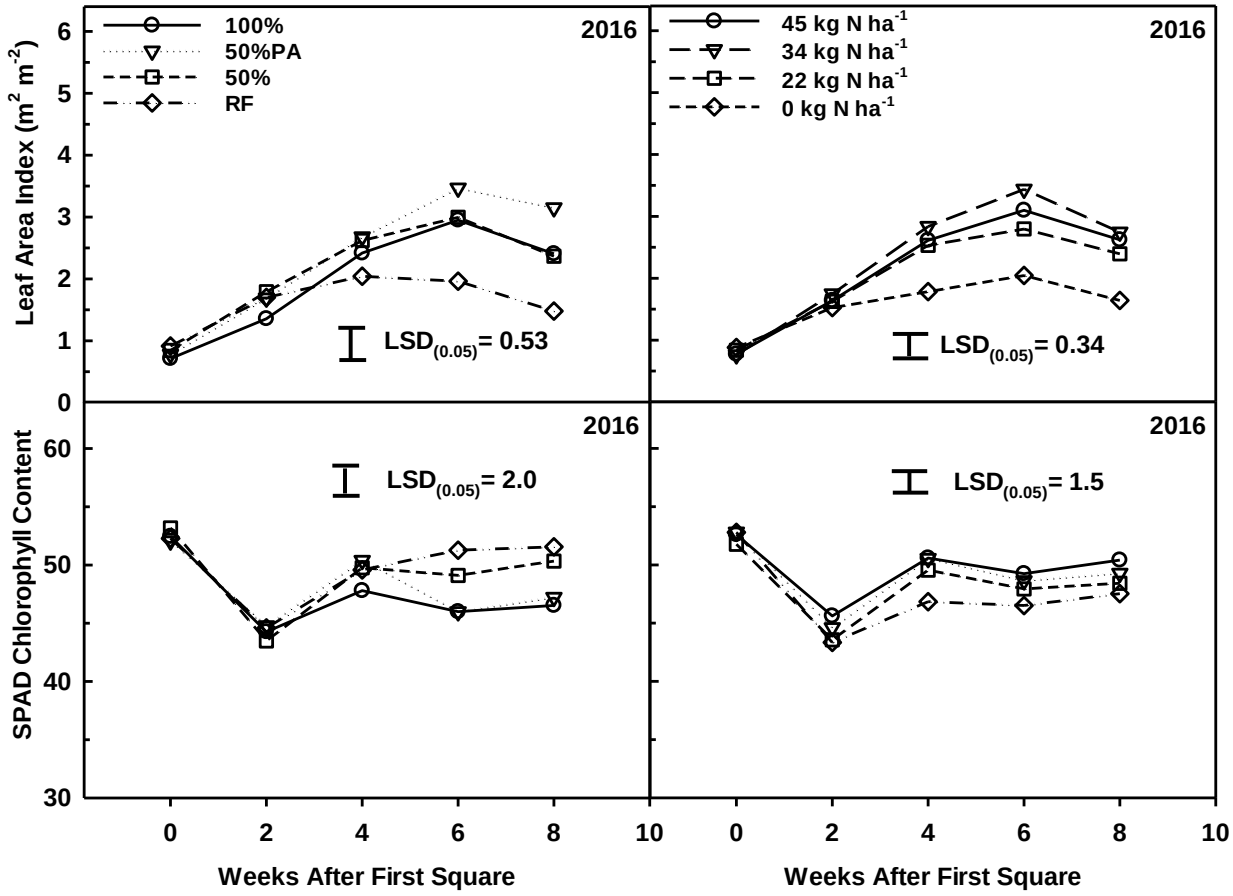

Figure 5-4. Top) Average leaf area index (LAI) starting at first square and measured until 8 weeks after first square for the irrigation and N treatments in 2016 at PSREU. Bottom) Average SPAD chlorophyll content starting at first square and measured until 8 weeks after first square for the irrigation and N treatments in 2016 at PSREU (Abbreviations: 100%, 100% PAWR per irrigation application; 50%PA, 50% PAWR per irrigation application until first flower followed by 100% PAWR per irrigation application; 50%, 50% PAWR per irrigation application for entire growing season; RF, rainfed control; Nitrogen rates are uniform in-season N rates applied at first square and bloom, first square, and first bloom; LSD, Fischer's Protected Least Significant Difference at $P < 0.05$).

Table 5-4. Average specific and phenomenological energy fluxes for each N rate in 2015 and 2016. Data is showing the in-season N rate, time within season, and year interaction. Parameters are averaged across both irrigation treatment and measurement date within a season. Measurements averaged across dates included sampling which occurred from first developed boll to first open boll.

Year	OJIP Parameters								
2015									
Nitrogen rate	J^{abs}/RC	J^{abs}/CS_o	J_o^{DI}/RC	J_o^{DI}/CS	J_o^{TR}/RC	J_o^{TR}/CS	J_o^{RE1}/RC	J_o^{RE1}/CS	RC/CS
0 kg N ha^{-1}	1.53	744	0.33	158	1.20	586	0.38	182	497
22 kg N ha^{-1}	1.52	761	0.33	162	1.19	599	0.38	191	513
34 kg N ha^{-1}	1.48	757	0.32	161	1.16	596	0.39	197	525
45 kg N ha^{-1}	1.46	764	0.31	160	1.15	603	0.41	214	534
$LSD_{(0.05)}$	0.06	13.8	0.02	3.84	0.04	12.4	0.02	9.52	19.7
2016									
Nitrogen rate									
0 kg N ha^{-1}	1.71	722	0.33	140	1.38	582	0.52	220	432
22 kg N ha^{-1}	1.68	728	0.33	143	1.34	584	0.50	217	445
34 kg N ha^{-1}	1.68	721	0.33	143	1.35	579	0.50	213	435
45 kg N ha^{-1}	1.72	719	0.34	142	1.38	577	0.52	217	425
$LSD_{(0.05)}$	NS	NS	NS	NS	NS	NS	0.02	NS	NS

† Nitrogen rates are uniform in-season rates split at first square and first bloom.

‡Abbreviations: J^{abs}/RC , average absorbed photon flux per PSII reaction center; J^{abs}/CS_o , average absorbed photon flux per cross section; J_o^{DI}/RC, dissipated energy flux in processes other than trapping per PSII reaction center; J_o^{DI}/CS, dissipated energy flux in processes other than trapping per cross section; J_o^{TR}/RC, maximum trapped exciton flux per PSII reaction center; J_o^{TR}/CS, maximum trapped exciton flux per cross section; J_o^{RE1}/RC, electron transport flux until PSI acceptors per PSII reaction center; J_o^{RE1}/CS, electron transport flux until PSI acceptors per cross section; RC/CS, number of active PSII reaction centers per cross section; LSD, Fischer's Protected Least Significant Difference at $P < 0.05$.

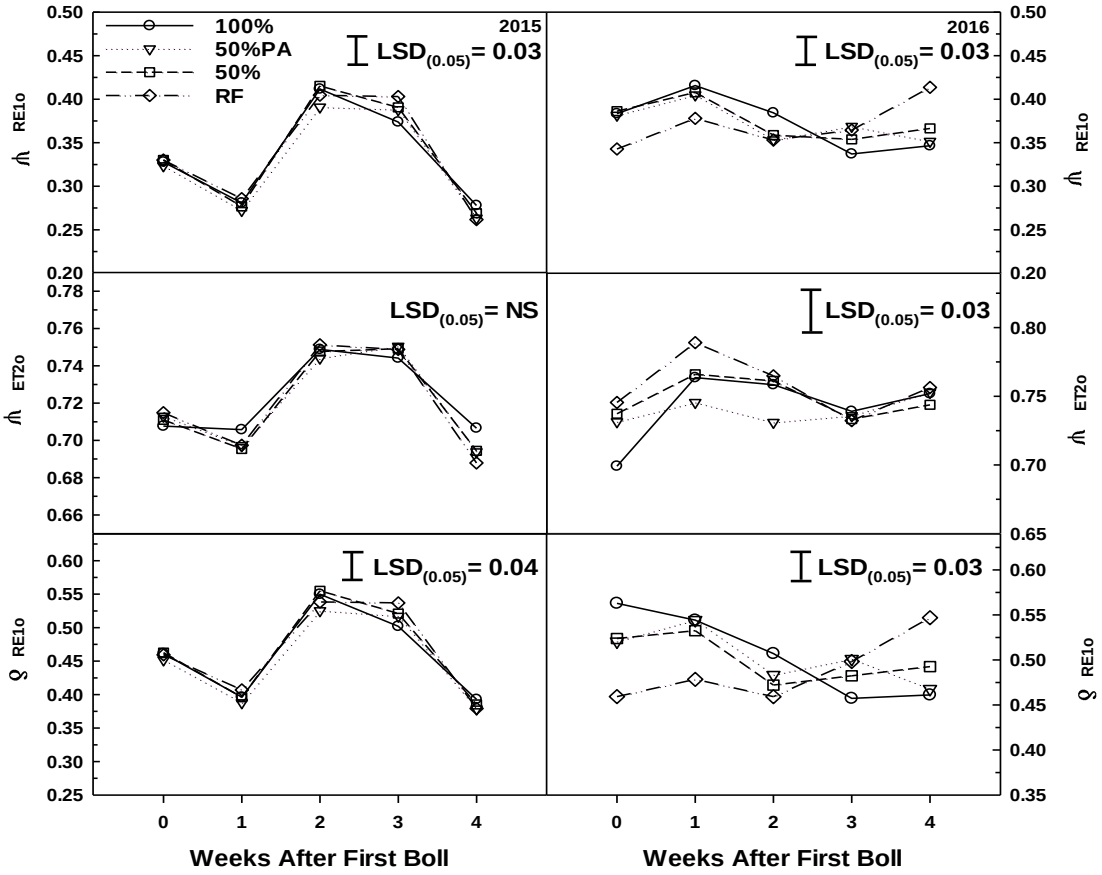

Figure 5-5. Electron transport efficiencies from first developed boll to first open boll in 2015 (left) and 2016 (right) (Abbreviations: Abbreviations: 100%, 100% PAWR per irrigation application; 50%PA, 50% PAWR per irrigation application until first flower followed by 100% PAWR per irrigation application; 50%, 50% PAWR per irrigation application for entire growing season; RF, rainfed control; ψ_{RE1o}, efficiency that a PSII trapped electron is transferred until PSI acceptors; ψ_{ET2o}, efficiency that a PSII trapped electron is transferred from Q_a to $Q_{b;}$ $\delta_{RE1o,}$ efficiency that a PSII trapped electron is transferred from Q_b to PSI acceptors; LSD, Fischer's Protected Least Significant Difference at $P < 0.05$).

Table 5-5. Analysis of variance (ANOVA) for N uptake, harvest index, and lint yield at the PSREU near Citra, FL.

Effect	N Uptake	Harvest Index	Lint Yield
year	0.0012	0.0748	0.4268
IR	0.4703	0.0846	<0.0001
year*IR	0.3373	0.3222	0.0135
N	<0.0001	<0.0001	0.3395
year*N	0.5407	0.2590	0.0002
IR*N	0.1102	0.5501	0.6780
year*IR*N	0.2099	0.5639	0.1100

†Abbreviations: year, growing seasons of 2015 and 2016 at the PSREU; IR, irrigation treatment; N , in-season N treatment.

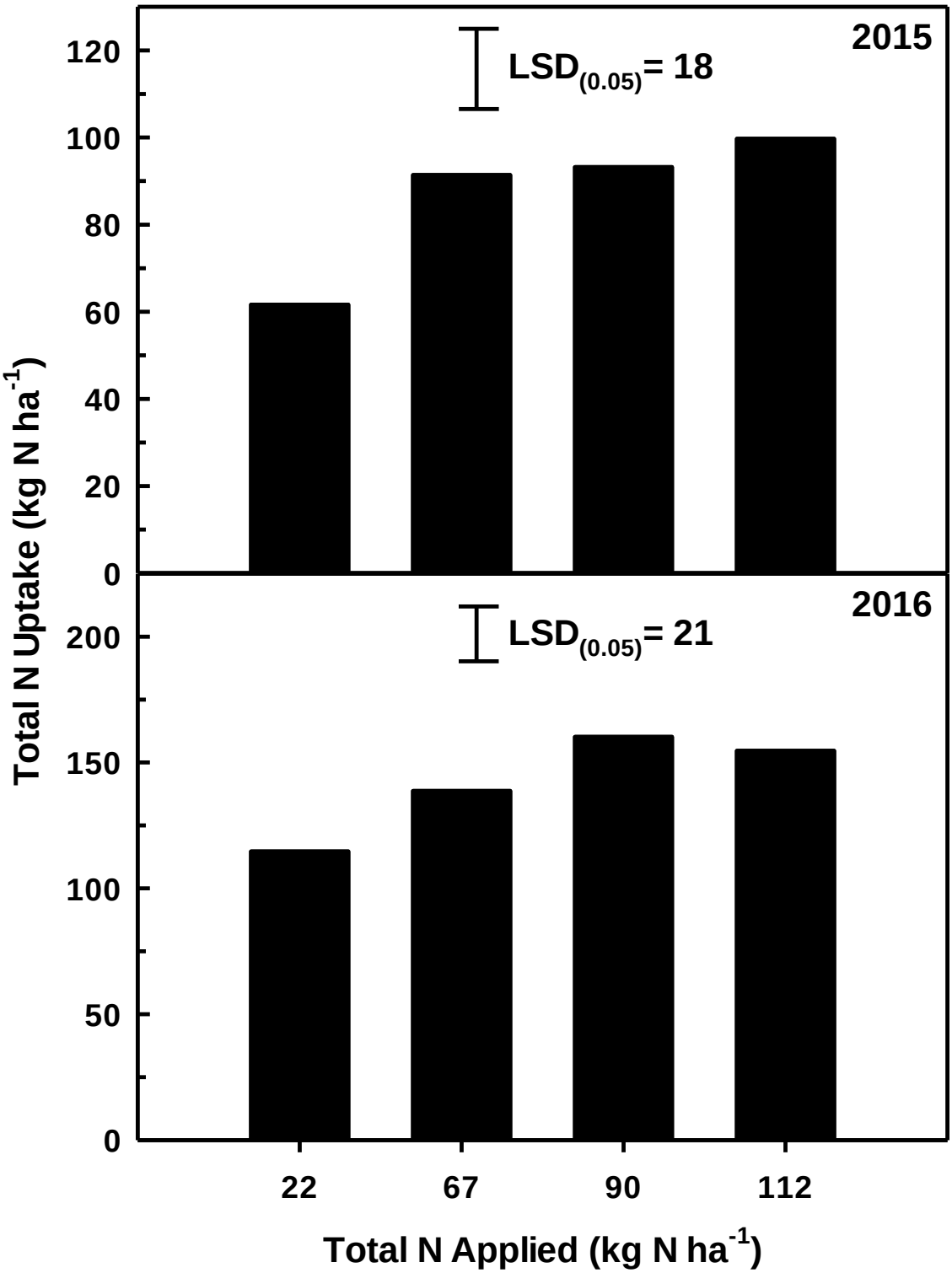

Figure 5-6. Average total N uptake for each N treatment in 2015 (top) and 2016 (bottom) at the PSREU near Citra, FL. Nitrogen rates are uniform in-season rates split at first square and first bloom. Data is average over each irrigation treatment showing the N effect for each year (Abbreviations: LSD, Fischer's Protected Least Significant Difference at $P < 0.05$).

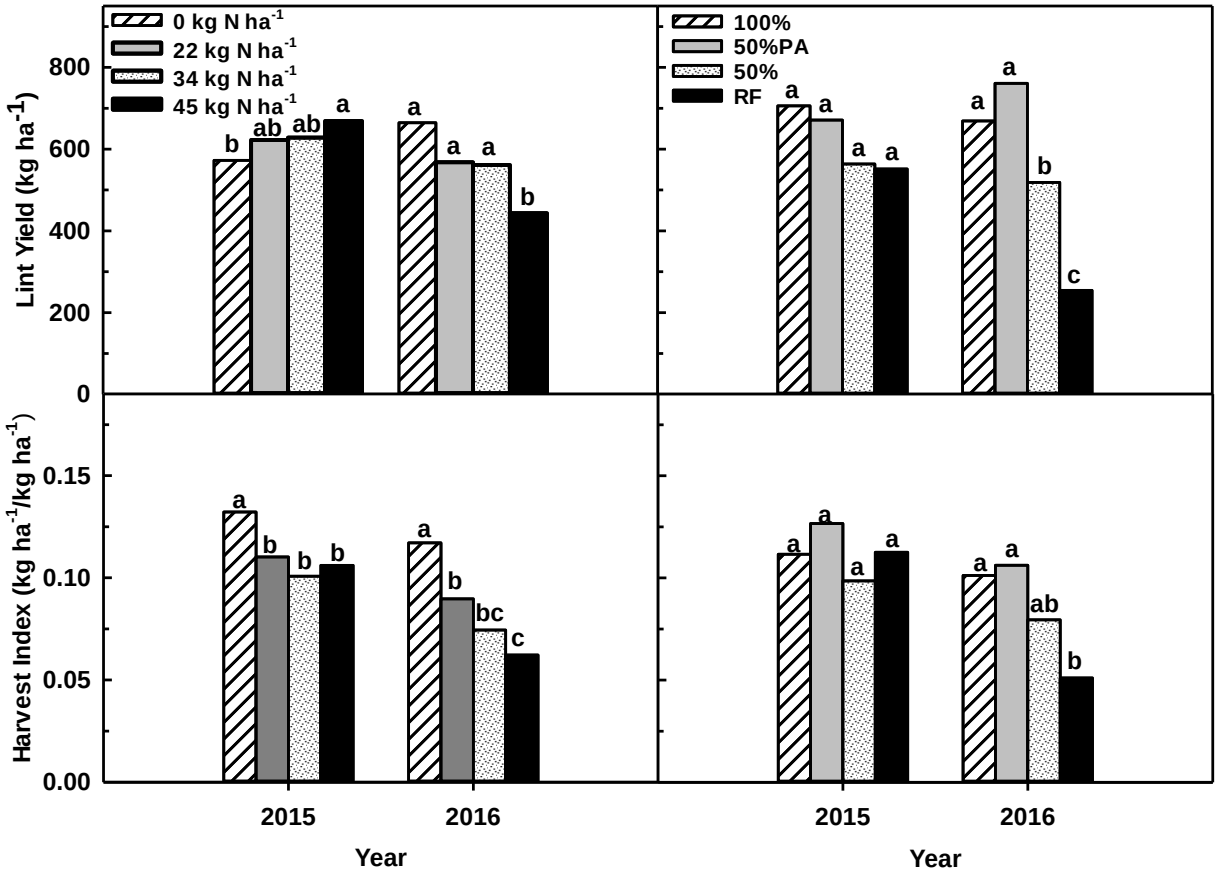

Figure 5-7. Average lint yield (top) and harvest index (bottom) for each N (left) and irrigation (right) treatment in 2015 and 2016 at the PSREU near Citra, FL. Different letters indicate significance using Fischer's Protected Least Significant Difference at $P < 0.05$ (Abbreviations: Nitrogen rates are uniform in-season rates split at first square and first bloom; 100%, 100% PAWR per irrigation application; 50%PA, 50% PAWR per irrigation application until first flower followed by 100% PAWR per irrigation application; 50%, 50% PAWR per irrigation application for entire growing season; RF, rainfed control).

CHAPTER 6
FINAL SUMMARY

A primary objective of the research presented in this dissertation was to determine the degree of pod yield stability across U.S. production regions and their relative response to irrigation within each environment. This objective was addressed in Chapter 2 by a pod yield stability analysis performed by examining the linear relationship between the average genotypic pod yield across all site years and irrigation treatments versus the population mean for each individual site year and irrigation treatment. Regression coefficients (b) of this linear relationship defining genotypic pod yield stability ranged from 0.88 to 1.30. Stable ($b \leq 0.90$) genotypes having broad range adaptability were C76 16, ICGS 76, Chico, TMV 2, and ICGV 86388; while unstable ($b \geq 1.10$) genotypes having specific adaptability were COC 041, Serenut 6T, and Serenut 5R. A genotype by irrigation interaction occurred at the Florida location, consisting of genotypes New Mexico Valencia C (NMVC), COC 041, and Chico responding positively to additional water applied, whereas, FloRun^{TM} '107', C76 16, and FlavorRunner 458 responded negatively. These results demonstrate both broad and specific adaptability of disparate peanut genotypes to U.S. production regions which could possibly be utilized by breeding programs for cultivar development.

Variation of peanut germplasm pod yield stability across environments, and in response to irrigation at the Florida location indicated diversity in genotypic phenotypes which may be contributing to these responses. This hypothesis was further evaluated in Chapter 3 of this dissertation by evaluating the above- and belowground traits of both genotypes which responded positively to increased water received and those which had no response. Genotypic total root length (TRL) and leaf area index (LAI) did not interact

with irrigation treatments, but decreasing the total amount of irrigation over the growing season reduced LAI and pod yield, with no impact on TRL growth or distribution to 80 cm of soil depth. Genotypic effects influenced the TRL development over the growing season, and genotypes of subspecies *fastigiata* had greater TRL deeper in the soil profile. However, all genotypes had similar amounts of total surface area (TSA) distribution to 80 cm of soil depth. A positive relationship was observed between pod yield and maximal LAI in both study years, although this significant relationship had a low correlation coefficient (r=0.39) in 2016 when greater water stress severity occurred during reproductive growth. A significant low negative correlation coefficient of -0.41 and -0.46 was observed between pod yield and maximal TRL in 2015 and 2016, respectively. The lack of interaction between irrigation and genotype for pod yield demonstrates that peanut genotypes which have more prolific root growth at depth may not necessarily have an advantage for increased amounts of water acquisition and utilization that are translated into yield.

The results in the Chapter 3 study were interesting in that they went against some of the reports in the literature which emphasize that a deep root trait is superior for improving water deficit stress resiliency. This lead to the hypothesis that simply more root presence, particularly deep in the soil profile may not necessary lead to greater crop water extraction. The research conducted in Chapter 4 set out to answer this question. A method was developed due examine the relationship between root architecture and soil water depletion distribution. The result of this experiment demonstrated that despite variations in genotypic RSA and responses to early season irrigation management, soil water depletion zones among the different RSA where

similar for each sample date. However, the soil water depletion zones interacted with sample date indicating that volumetric soil water content mostly impacted where the soil water depletion was occurring in the soil profile. When soil water content was high following irrigation, approximately 74% of the quantified soil water depletion occurred in the top 30 cm of soil. Under soil drying conditions, the predominate proportion of soil water depletion occurred at 30-60 cm of soil depth accounting for 67% of the total soil water uptake. The control of hydrologic conditions on soil water depletion zones is likely why no significant relationship were observed between total root volume (TRV) and total soil water depletion. Furthermore, comparing daily volumetric soil water content among the two genotypes to mid-morning transpiration fluxes provided evidence of differences in genotypic diurnal hydraulic conductivities. Overall, these results suggest that trait selection for improving utilization in the humid southeast irrigated peanut production environment may consist of genotypes which have a greater proportion of its root density in surficial soils to capture in-season water inputs, can obtain high maximal daily transpiration rates, but are sensitive to decreasing transpiration rates one daily increases in vapor pressure deficit (VPD) occur.

The final objective of the research in this dissertation was to determine optimal irrigation and in-season N management strategies for the southeast cotton production system. Optimal irrigation management was determined by evaluate varying levels of plant available water replacement (PAWR) estimated by the ET-based soil water balance model Cotton SmartIrrigation smartphone application. A strength of using this method is that this irrigation scheduling tool makes recommendations based on site specific environmental. Two years of analysis over a wet and dry year indicated that

optimal irrigation management for this soil types was to apply 60% PAWR up until first bloom with subsequent irrigation amount being 100% PAWR. Both years of this study also showed that lint yield maintenance or gains occurred with making two split N applications at first square and bloom of 22 kg N ha^{-1}, a 45 kg N ha^{-1} reduction in comparison to the traditional total N recommendation of 112 kg N ha^{-1}.

The knowledge gained from this research has potential to improve peanut and crop production as a whole. This research has identified specific peanut genotypes which have relatively high yield and stability across production regions, and identified inherent biomass partitioning differences among peanut genotypes of varying taxonomic descent. Both of these contributions have potential to assist breeders in peanut cultivar development. Another impact his research had is the development of a phenotyping protocol to examine the coordination of pertinent traits which can be used to determine specific adaptability of crop genotypes to climates and production regions of varying soil hydrological conditions. Overall these contributions have potential to improve the adaptability of crops, which when combined with site-specific management support tools have the potential to improve yield production, grower profitability, and environmental sustainability.

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BIOGRAPHICAL SKETCH

Brendan Zurweller is a native of Missouri where he received his Bachelor of Science in Environmental Science at the University of Missouri-Columbia. During his undergraduate education he had a strong interest in soil science which he applied during his graduate research at the University of Missouri-Columbia. His research during this appointment was focused on examining the impacts of N management and soil waterlogging on both greenhouse gas emissions and corn grain production. During this research he developed an interest for crop responses to abiotic stress. This led him to pursue a doctoral degree with research encompassing crop trait responses to water and nitrogen management. Upon completing his doctoral degree, Brendan hopes to continue in a career where he can continue conducting agronomic related research.