PHYSIOLOGICAL CONSEQUENCES OF LATE LEAF SPOT ON PEANUT (*Arachis hypogaea* L.) CULTIVARS OF DIFFERING RESISTANCE

By

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To my mom

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LIST OF ABBREVIATIONS

A_{sat}	Light-saturated leaf CO_2 assimilation rate
ANOVA	Analysis of Variance
DAP	Days After Planting
ELS	Early Leaf Spot (caused by *Cercospora arachidicola*)
F_v/F_m	Maximum efficiency of PSII photochemistry after dark-adaptation
LLS	Late Leaf Spot (caused by *Cercosporidium personatum*)
PPFD	Photosynthetic Photon Flux Density
stAUDPC	Standardized Area Under the Disease Progress Curve
TCP	Total Canopy Photosynthesis
TSMK	Total Sound Mature Kernels
TSWV	Tomato Spotted Wilt Virus
$V_{c,max}$	Maximum carboxylation velocity of Rubisco
ϕ_{CO2}	Quantum efficiency of CO_2 assimilation

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In the southeastern United States, late leaf spot (LLS) caused by *Cercosporidium personatum* is one of the most widespread foliar diseases, which can substantially reduce peanut yields, unless controlled by regular and costly fungicide applications. Crop protection strategies based on minimizing crop losses by studying host response to the pathogen rather than minimizing disease outbreaks offer a promising way to reduce fungicide use and improve cultivar selection procedures. The overall goal of this study was to characterize LLS severity and progression and its impact on growth, yield and photosynthetic metabolism of peanut cultivars with differing levels of resistance to LLS. Field experiments were conducted over two years at Citra, FL evaluating two peanut cultivars with more (York) and less (Carver) quantitative resistance to LLS, grown under fungicide sprayed and non-sprayed conditions. The first objective of this study was to quantify the effects of LLS on growth and yield in peanut cultivars of differing resistance. Disease severity based on canopy lesion area was reduced by 30% in York compared to Carver. No additive effects of combining the resistant cultivar with fungicide were seen, as fungicide use increased yield by 364 kg ha^{-1} for both cultivars. Yield was more strongly related to disease severity based on canopy lesion area than to

the Florida scale. Despite reduced disease severity, pod yield gain was only 6% in York compared to Carver. The second objective of this study was to quantify the effects of LLS on leaf photosynthetic traits in peanut cultivars. To analyze the leaf photosynthesis (A_{sat}) data, a non-linear model, $y = (1 - x)^{\beta}$ was used, where y is relative A_{sat}, x is measured visual lesion area, and β represents the relationship between virtual and visual lesion area. Progression of LLS severity on leaf cohorts was slower in York compared to Carver. However, the reduction in A_{sat} with leaf cohort age was similar across the cultivars. This paradox could be explained by a higher β value in York (4.6) compared to Carver (3.6), indicating a greater relative reduction in A_{sat} beyond the necrotic lesion area in York. This greater reduction in A_{sat} in York compared to Carver was most closely related to a reduction in maximum carboxylation velocity and chlorophyll. The third objective of this study was to simulate growth and yield as affected by LLS on peanut cultivars of differing resistance. The CROPGRO-Peanut model was able to simulate the observed leaf, pod, and total dry biomass over time when inputs on percent necrotic leaf area and defoliation were provided. Correlations among measured defoliation and necrotic leaf area with visual disease ratings indicated that visual disease ratings could be successfully used to estimate necrosis and defoliation and to correctly simulate LLS induced reductions in growth and yield. Results from this study indicated that future efforts to improve LLS resistance should include sustaining A_{sat} (i.e. lower β value) under LLS infection along with slower disease progress.

CHAPTER 1
INTRODUCTION

Peanut (*Arachis hypogaea* L.) is one of the major sources of protein and oil in the world. It is cultivated on 24 million hectares in over 100 countries, generating an annual production of nearly 37 Tg (FAO, 2011). Nevertheless, worldwide peanut production is severely hampered by the incidence of numerous diseases. Early leaf spot (caused by *Cercospora arachidicola* S. Hori), and late leaf spot [caused by *Cercosporidium personatum* (Berk. & Curt.) Deighton] are among the most widespread and damaging foliar diseases of peanut throughout the world (McDonald et al., 1985). Pod yield losses of up to 50% are common when fungicides are not applied. These losses may reach as high as 70% when disease is not controlled (Shokes and Culbreath, 1997). In Florida, late leaf spot (LLS) is the predominant foliar disease (Jackson, 1981), causing yield losses of up to 50% (Pixley et al., 1990a).

Consequently, regular and costly fungicide applications are currently used to minimize yield losses from peanut diseases (Woodward et al., 2008; Monfort et al., 2004). Improved cultivars with moderate resistance to late leaf spot, along with other integrated disease management practices, have also been successfully used to reduce inputs and production costs (Woodward et al., 2010; Woodward et al., 2008; Monfort et al., 2004). However, the effects of LLS on the physiological responses in cultivars of differing leaf spot resistance is not well understood and could contribute to better identification of improved cultivars in breeding programs and better modeling estimates of peanut yield loss to LLS. Moreover, economic and environmental concerns of fungicide use have increased the demand for improved management strategies based on minimizing crop losses rather than minimizing disease outbreaks. Decision support

systems that can predict yield losses rather than controlling the outbreak of disease and hence provide improved disease management options are required to meet this demand.

Late leaf spot first occurs as necrotic lesions on peanut leaflets and subsequently induces leaflet abscission. Premature loss of green leaf area (by necrotic tissue and defoliation) and reduction of leaf photosynthetic capacity due to LLS contribute to a loss of canopy carbon assimilation, and thus a loss of yield. Many older peanut cultivars such as Florunner and Georgia Green have poor resistance to LLS. Loss of leaf area due to accelerated senescence was reported to be the predominant mechanism of yield losses in those cultivars (Bourgeois and Boote, 1992; Boote et al., 1980). However, for cultivars with improved resistance to LLS that experience less defoliation, yield reduction may be affected to a greater extent by the leaf physiological response to disease rather than primarily to loss of leaf area.

Fungal pathogens generally reduce leaf photosynthesis not only through a reduction in green leaf area, but also through an effect on photosynthetic capacity of the remaining green leaf tissue (Bastiaans, 1991). In order to relate reductions in leaf photosynthesis to visual lesion area, Bastiaans (1991) proposed a relatively simple model, $y = (1- x)^{\beta}$, where y is the relative net assimilation rate of a diseased leaf compared to that of an asymptomatic leaf, x is the measured visual lesion area, and β describes the relationship between virtual and visual lesion area. The virtual area represents loss of photosynthetic capacity beyond the visual lesion area. Thus, β indicates whether the effect of disease on photosynthesis is higher ($\beta > 1$), lower ($\beta < 1$), or equal ($\beta = 1$) to that accounted for by the measured visual lesion area. Using this

model, several studies have shown that the reduction in photosynthesis occurred beyond the measured lesion area (Bassanezi et al., 2002; Erickson et al., 2003; Zhang et al., 2009). Understanding why *β* values differ, and perhaps even more importantly, whether they differ by cultivar within species is critical for improved cultivar selection and for modeling effects of disease on carbon assimilation, growth and yield (Bastiaans, 1993; Adomou et al., 2005; Bancal et al., 2007). Some studies have reported variation in *β* value within cultivars of the same species (Erickson et al., 2003; Zhang et al., 2009). Despite the importance of LLS in peanut production, there are no published *β* values for newly released cultivars or comparisons among peanut cultivars.

Crop models are essential tools to evaluate growth and yield losses due to various biotic and abiotic stresses (Boote et al., 1983a; Naab et al., 2004; Timsina et al., 2007). Models used to predict the impact of foliar diseases on yield have generally incorporated the disease effects on defoliation and photosynthesis (Batchelor et al., 1993; Teng et al., 1998). Quantification of the effect of LLS on photosynthetic metabolism of peanut cultivars with variable resistance levels to LLS and its inclusion in yield loss models is of great importance for a more complete understanding of growth and yield responses to diseases and improved accuracy of crop models. The CROPGRO-Peanut model (Boote et al., 1998a, 1998b) is a process-oriented mechanistic crop growth model which considers crop carbon balance, crop and soil N balance, and soil water balance at the process level. This model has coupling points and procedures for entering pest damage to simulate growth and yield reductions associated with foliar pathogens like LLS (Batchelor et al., 1993; Boote et al., 1993). The primary impacts of disease are simulated as defoliation; however the impacts of

virtual lesion area are currently simulated by simply defoliating more leaf area rather than creating direct impact at the leaf level photosynthesis. This subroutine has not been subjected to rigorous testing as previous studies testing this subroutine did not have measured data on necrosis and/or defoliation (Naab et al., 2004; Adomou et al., 2005). Further, modification may be warranted in the model to directly impact the leaf level photosynthetic metabolism as observed in the field (Bastiaans, 1991; Bourgeois and Boote; 1992) rather than defoliating more leaf area.

The overall goal of this study was to characterize variability in LLS severity and progression and its impact on growth, yield and photosynthetic metabolism of peanut cultivars with variable levels of resistance to LLS. More specifically, this study was conducted with the following objectives:

- To characterize LLS severity and its effects on growth and partitioning, leaf lifespan, canopy photosynthesis, and pod yield of York, a relatively resistant cultivar, compared to Carver, a cultivar with relatively poor resistance to LLS in a field environment.
- To quantify the effects of LLS on leaf photosynthetic metabolism in two peanut cultivars with variable levels of resistance to LLS.
- To evaluate the CROPGRO-Peanut model for its ability to simulate the impacts of LLS on growth and yield reductions in peanut cultivars with differing resistance levels with measured inputs of necrosis and defoliation.

CHAPTER 2
LITERATURE REVIEW

Background

Peanut is a legume grown in warm climates throughout the world. It is cultivated around the world in tropical, sub-tropical, and warm temperate climates. The cultivated peanut (*Arachis hypogaea* L.) originated in South America and is a self-pollinating, indeterminate, annual plant that is distinguished from most other species by producing aerial flowers but fruiting below the soil surface. Peanut leaves, the photosynthetic unit of the plant, are pinnate with two opposite pairs of leaflets. The primary center of diversity for the species is the Chaco region between southern Bolivia and northwest Argentina (Gregory and Gregory, 1979).

Peanut is now cultivated on 24 million ha in more than 100 countries between 40^{o} N and 40^{o} S, generating an annual production of nearly 37 Tg (FAO, 2011). During 2009, China, India, Indonesia, and United States (US) accounted for almost 70% of the total world production, with China leading the way at 40% of production. Average pod yield ranged from 1000 kg ha^{-1} in India to 3800 kg ha^{-1} in US. In the US, peanut was produced on about 0.52 million ha during 2010, with a total production of 1.88 Tg. The US peanut production is concentrated mainly in three major geographic areas: (i) the southeast, which includes Georgia, Alabama, Florida, and Mississippi; (ii) the southwest, which includes Texas, New Mexico, and Oklahoma; and (iii) Virginia-Carolina, which includes North Carolina, South Carolina, and Virginia. In 2010, Georgia had the highest area and production (228,647 ha and 0.90 Tg) followed by Texas (66,773 ha and 0.27 Tg), Alabama (76,890 ha and 0.22 Tg), and Florida (58,679 ha and 0.21 Tg). The southeastern US accounted for 77% of total US production. Florida

provides about 11% of US peanut production valued at around $91 million (USDA NASS, 2011).

A. *hypogaea* is divided into two subspecies (*hypogaea* and *fastigiata*) and six botanical varieties (Krapovickas and Gregory, 1994). The subspecies *hypogaea* which includes botanical varieties *hypogaea* and *hirsuta* does not flower on the main stem, has alternate branching pattern, is late maturing, has a high water requirement, and produces larger seeds. The subspecies *fastigiata* which includes botanical varieties *fastigiata*, *peruviana*, *aequatoriana*, and *vulgaris* produces flowers on the main stem, has sequential branching, matures earlier, has a low water requirement, and produces smaller seeds.

The four market types in the US peanut trade are: Runner, Virginia, Spanish, and Valencia. Botanical variety *hypogaea* contains the Virginia and Runner market types, *fastigiata* contains the Valencia market types, and *vulgaris* contains the Spanish types. These market types form a rough classification system based on pod and seed size characteristics and to a lesser extent on center of genetic origin, growing region, and growth habit (Knauft et al., 1987).

Spanish market types typically have small kernels covered with a reddish-brown skin. They are used primarily in candies and crushed for oil. They are grown mostly in Texas and Oklahoma. Valencia market types have multi-kernel pod characteristics, red seed coats, and medium seed size. They are grown primarily in the southeastern US and usually used for boiling and roasting. Runner market types tend to have larger pods and seeds compared to Spanish and Valencia types. They are most widely grown in the southeastern US growing region and are used for oil and peanut butter production.

Virginia market types have a large pod and seed size. They are primarily grown in North Carolina, South Carolina, and Virginia and used in snack nuts.

Peanut Diseases

Peanut is susceptible to a variety of biotic stresses. The development and severity of peanut diseases depends on complex interactions among the host, the pathogen, and the environment. The most prevalent pathogens of peanut include tomato spotted wilt virus (TSWV, Topsovirus vectored by thrips), *Sclerotium rolfsii* Sacc., the causal agent of white mold, *Sclerotinia minor* Jagger, the causal agent of Sclerotinia blight, *Cylindrocladium parasiticum* Crous, Wingfield and Alfenas, the causal agent of Cylindrocladium black rot, *Puccinia arachidis* Speg., the causal agent of rust, and *Cercospora arachidicola* S. Hori and *Cercosporidium personatum* (Berk and Curt.) Deighton, the causal agents of early leaf spot (ELS) and late leaf spot (LLS). Many of these diseases of peanut have a limited geographic range, but the two major foliar diseases, early and late leaf spot are prevalent in almost all peanut producing regions of the world (McDonald et al., 1985; Stalker, 1997) and result in yield reductions that may approach 70% in the absence of proper management practices (Nutter, Jr. and Shokes, 1995; Shokes and Culbreath, 1997). In Florida, previous research has indicated that LLS is the predominant disease (Jackson 1981; Pixley et al., 1990a, 1990b; Nutter Jr. and Shokes, 1995).

Late Leaf Spot

Late leaf spot caused by *Cercosporidium personatum* (Berk. & Curt.) Deighton, (telemorph = *Mycosphaerella berkeleyi* Jenk.) is the most widespread foliar disease of peanut in the southeastern US (Jackson, 1981). Pod yield losses can be up to 70% when fungicides are not applied (Shokes and Culbreath, 1997; Pixley et al., 1990a,

McDonald et al., 1985; Jackson and Bell, 1969). The anamorph *C. personatum* is commonly observed on peanut. The lesion is amphigenous with fruiting on both sides of the leaflet, but sporulation is more common on the lower (abaxial) surface. The telemorph, *M. berkeleyi* is rarely observed on peanut. Conidiophores of *C. personatum* form in dense clusters, are pale to olivaceous brown, have conspicuous conidial scars, and vary in size from 10-100 μ x 3.0-6.5 μ. Conidia are medium olivaceous, cylindrical, obclavate, usually straight or slightly curved and have 1-9 (mostly 3-4) septa. They vary in size from 20-70 μ x 4-9 μ (McDonald et al., 1985). The host range of *C. personatum* is confined to the genus *Arachis*.

Symptoms

Although the disease is called LLS, the symptoms of this disease develop on petioles, stipules, stems, and even pegs during the later stages of an epidemic. Lesions are first visible around 10 days after spore deposition as tiny, pinpoint, yellowish flecks. These flecks enlarge to form coalescing, blackish-brown lesions, enlarging to 1-10 mm in diameter (Shokes and Culbreath, 1997). Mature, sporulating lesions may be apparent by about 15 days after spores are deposited. Sporulation occurs mainly on the lower leaflet surface, although some spores may also be produced on the upper surface of older lesions (Jenkins 1938). Late leaf spot lesions commonly have a less conspicuous or absent chlorotic halo.

Disease Cycle

Conidia, produced by conidiophores on peanut residues in the soil and off-season plants serve as a source of initial inoculum (Figure 2-1). Although the telemorph of the fungus is known, the ascospores are generally not regarded as important source of

primary inoculum. Conidia are dispersed by wind, splashing water, and insects. Peak dispersal period for conidia occurs at dew dry-off in the morning and at the onset of rainfall. Multi-celled conidia land on peanut tissue and start germinating. Pathogenesis of *C*. *personatum* starts with the development of spore germination tubes which enter plant cells via stomata (Leal-Bertioli et al., 2010) or directly through epidermis, allowing intercellular mycelium growth. *Cercosporidium personatum* does not kill the host cells prior to penetration, but instead develops into haustoria (Perfect and Green, 2001; Mims et al., 1988; Abdou et al., 1974). Lesions generally develop within 10-14 days of initial infection (Shokes and Culbreath, 1997). In spite of having a longer incubation period, LLS fungus produces more spores per lesions compared to ELS fungus resulting in more severe damage over a short period of time. Variables like weather, cultivar, and the effectiveness of control measures determine number of disease cycles.

Cercosporidium personatum favors warm temperature and humid conditions, which are found in the southeastern peanut growing states during summer months. Maximum LLS infection occurs when temperatures are about 20^{o}C and relative humidity exceeds 93% for more than 12 hr or with continuous leaf wetness periods of 10 hr (Shokes and Culbreath, 1997). Prolonged periods of leaf wetness or several shorter periods of leaf wetness (10 hr or longer) may be equally favorable for LLS development. Shew et al. (1988) reported that infection diminished with increase in temperature from 20^{o}C (where maximum infection occurred) to 28 to 32^{o}C. Leaf wetness is an important limiting factor for infection (Butler et al., 1995).

Management Strategies

Management of late leaf spot of peanut has been accomplished through the combination of (i) reducing initial inoculum by crop rotation, early planting, and tillage,

and (ii) reducing the rate of disease spread by cultivar selection and multiple applications of fungicides. Late leaf spot is much worse in warm and wet weather than in cool and dry weather, making it more difficult to control.

Cultural Practices

Most cultural practices used to control LLS are aimed at reducing the initial inoculum. These include crop rotation, early planting, tillage, burial of crop residues, and removal of volunteer peanut plants. However, these methods alone will not permit sustained peanut production (Kucharek, 2005).

Crop rotation

Crop rotation is one of the most effective means of managing disease in any crop. It results in increased effectiveness of all disease management programs. When peanut is planted on land which has not been planted to peanut for 3-4 years, the onset and progress rate of LLS are delayed, as contrasted with continuous peanut culture. This alone might provide a 2-3 week delay in the development of LLS epidemic. It is recommended to avoid planting peanut in the same field more than once out of every three, preferably four years. Considering all peanut diseases, it is recommended to rotate peanut with grass crops such as corn, sorghum, and bahiagrass (Kemerait et al., 2010; Mossler and Aerts, 2007). Peanut yields were reported to be 19 and 41% higher after two years of corn and two years of bahiagrass, respectively (Wright et al., 2006).

Planting date

Early planting dates (early to mid-April) result in comparatively less exposure time for peanut canopies to hot and humid conditions most conducive for LLS development. This results in peanut canopies exposed to LLS for less period before harvest (Shokes et al., 1982). Because inoculum concentration increases as the season progresses, LLS

can be managed to some degree by manipulating planting dates. Mossler and Aerts (2007) reported that peanut planted in early- to mid-April in Florida may not have to be sprayed with fungicides until 60 days after planting (DAP), compared to 25-35 DAP for peanut planted during May to early June. However, this advantage is negated by more susceptibility of earlier planted peanut to TSWV (Kemerait et al., 2010).

Tillage

Conventional tillage involves turning the soil in an entire field, resulting in incorporation of plant residues into the soil. Enhanced residue decay after tillage favors nonpathogenic soil micro-organisms over pathogens, resulting in reduction of overwintering inoculum and better disease control (Nutter, Jr. and Shokes, 1997). Recent interests in conservation tillage (e.g. strip tillage) due to increased energy and labor costs prompted studies evaluating effects of conservation tillage on disease epidemics. Although the exact mechanism is not clear, LLS epidemics in strip-tilled peanut fields were similar or suppressed compared with conventional tilled plots (Monfort et al., 2004; Cantonwine et al., 2006; Wright et al., 2006; Kemerait et al., 2010).

Fungicide Application

Multiple applications of fungicides are usually required to keep LLS disease below damaging levels. Fungicide application currently accounts for one-third of the total variable costs needed to produce peanut (Mossler and Aerts, 2007). Commonly used foliar fungicides include chlorothalonil, tebuconazole, propiconazole, pyraclostrobin, azoxystrobin, trifloxystrobin, and sulfur. Less commonly used fungicides include copper, maneb, mancozeb, and thiophanate (Mossler and Aerts, 2007). Fungicides are currently applied beginning approximately 30-40 DAP and continuing at 10-14 day intervals

resulting in seven or more applications during a growing season (Wright et al., 2006; Kemerait et al., 2010). Shokes et al. (1982) reported reduced disease severity and defoliation and higher yields with earlier initiation of fungicide application in Florida. Many other studies have shown the use of fungicide application in control of LLS (Smith and Littrell., 1980; Shokes et al., 1983; Bourgeois et al., 1991; Monfort et al., 2004; Woodward et al., 2008; Woodward et al., 2010).

Indiscriminate application of fungicides for LLS control may result in undesirable effects, e.g. development of fungicide-tolerant strains of fungus (Smith and Littrell., 1980) and increased severity of other diseases (Shokes and Culbreath, 1997). Several disease-forecasting systems have been developed based on relative humidity, temperature or simply number of rain events to reduce spray frequencies (e.g. AU-Pnut advisory, Jacobi et al., 1995; Jacobi and Backman, 1995). However, hot and humid weather in Florida result in limited use of these weather-based advisories. Moreover, implementation of these systems results in control of disease outbreak rather than minimizing yield losses.

Cultivar Selection

Cultivars partially resistant to *C. personatum* may also be used to reduce the rate of LLS epidemics. The highest levels of partial resistance are found in unadapted germplasm lines and in wild species-derived breeding lines (Wynne et al., 1991), resulting in slow progress in breeding for resistance to LLS. Identified components of rate-reducing partial resistance include extended latent period of the fungus, reduced sporulation, and smaller lesion diameters (Cook, 1981; Chiteka et al., 1988; Aquino et al., 1995; Dwivedi et al., 2002; Cantonwine et al., 2008). Mechanisms of resistance

include restriction of conidial development and penetration of fungal hyphae through stomata (Leal-Bertioli et al., 2010).

Breeding and selection of cultivars with partial resistance to LLS have been an important part of integrated disease management programs for reducing yield losses in peanut (Tillman et al., 2008; Tillman and Stalker, 2009). Several new releases have shown good resistance associated with delayed disease progress and decreased defoliation. These include Southern Runner (Gorbet et al., 1987), C-99R (Gorbet and Shokes, 2002a), Georgia-01R (Branch, 2002), Florida MDR 98 (Gorbet and Shokes, 2002b), Georgia-05E (Branch, 2006), Tifrunner (Holbrook and Culbreath, 2007), Hull (Gorbet, 2007), DP-1 (Gorbet and Tillman, 2008), Tifguard (Holbrook et al., 2008), Georgia-07W (Branch and Brenneman, 2008), Georganic (Holbrook and Culbreath, 2008), and York (Gorbet and Tillman, 2011). The degree of resistance in these cultivars is partial and still allows for significant damage under severe disease epidemics.

Several of the newly released cultivars are associated with unfavorable characteristics such as poor germination (e.g. York, DP-1, C-99R, and Hull) and late maturity (require extra 14-21 days). Poor seed emergence results in reduced field stands and hence lower final yields (Morton, 2007).

The best management strategy for LLS should integrate several of the above tactics into a program adapted to the cultivar and cultural practices of a given area.

Effects of Late Leaf Spot on Peanut Physiology

Peanut economic yield is a function of cumulative biomass and harvest index, which is determined by partitioning of assimilates to pod and effective duration of pod fill (Phakamas et al., 2008; Duncan et al., 1978). Late leaf spot disease first occurs as necrotic lesions on peanut leaflets and subsequently induces leaflet abscission. This

defoliation commonly lowers LAI values below the optimum value of 3.0 determined by Duncan et al. (1978) which reduces light interception and can cause significant loss of canopy carbon assimilation and yield (Bourgeois and Boote, 1992; Boote et al., 1983a). In addition to lowered LAI, reduction in carbon assimilation capacity of the leaves infected with LLS compared to the asymptomatic leaves has also been reported (Boote et al., 1980; Bourgeois and Boote, 1992). Thus, premature loss of green leaf area (by necrotic tissue and defoliation) and reduction of leaf photosynthetic capacity due to disease can contribute to a loss of canopy carbon assimilation, and thus a loss of yield.

Many older peanut cultivars such as Florunner and Georgia Green have poor resistance to LLS. Loss of leaf area due to accelerated senescence was reported to be the predominant mechanism of yield loss in these cultivars (Bourgeois and Boote, 1992; Boote et al., 1980). Bourgeois et al. (1991) reported a reduction of 37 and 46% in pod yield of Florunner in two seasons due to the loss of green photosynthetic leaf area causing significant reduction in production of carbohydrate available for pod growth.

However, for cultivars with improved resistance to LLS that experience less defoliation (Anderson et al., 1993; Knauft and Gorbet, 1990), yield reduction may also be related to the leaf physiological response to disease instead of to loss of leaf area alone. Late leaf spot disease is characterized by chlorotic flecks that enlarge to necrotic lesions that reduce photosynthetic capacity (Boote et al., 1983a). Necrotic lesions are photosynthetically useless area that does intercept light. However, there may be an additional effect on the photosynthetic capacity of non-infected symptomless area of the leaf.

The concept of a virtual lesion, introduced by Bastiaans (1991), can help in the classification of pathogens according to their effect on photosynthetic efficiency of their hosts. According to Bastiaans (1991), the virtual lesion is the proportion of leaf tissue, equal to or larger than the visual lesion (proportion of leaf tissue with visible symptoms), in which photosynthesis is severely reduced. In order to relate reductions in leaf photosynthesis to visual lesion area, Bastiaans (1991) proposed a relatively simple model, $y = (1- x)^{\beta}$, where y is the relative net assimilation rate of a diseased leaf compared to that of an asymptomatic leaf, x is the measured visual lesion area, and β describes the relationship between virtual and visual lesion area. The virtual area represents loss of photosynthetic capacity beyond the visual lesion area. Thus, β indicates whether the effect of disease on photosynthesis is higher ($\beta > 1$), lower ($\beta < 1$), or equal ($\beta = 1$) to that accounted for by the measured visual lesion area.

Using this model, several studies have shown that the relationship between photosynthesis and visual disease severity is related to host-pathogen interactions (Bassanezi et al., 2002; Erickson et al., 2003; Zhang et al., 2009). In general, studies have indicated that β values are relatively low (< 2.5) for biotrophic pathogens (Bassanezi et al., 2001; Lopes and Berger, 2001; Robert et al., 2005; Kumudini et al., 2010), intermediate (3 to 6) for hemibiotropic pathogens (Bassanezi et al., 2001; Erickson et al., 2003; Roloff et al., 2004) and highest for necrotrophic pathogens (Bassanezi et al., 2001; Lopes and Berger, 2001).

Values of β reported for some biotrophic pathosystems include 1.3 and 2.2 for *Uromyces appendiculatus* (Pers.:Pers) Unger on common bean (*Phaseolus vulgaris* L.) (Lopes and Berger, 2001 and Bassanezi et al., 2001, respectively) and 2.3 for

Phakospora pachyrhizi Syd. & P. Syd. on soybean (*Glycine max* L. Merr.) (Kumudini et al., 2010). Thus, *β* values of biotrophs have generally been equal to or very close to one, indicating minimal effects of biotrophic pathogens on photosynthesis beyond the visual lesion areas of the leaf. However, *β* values reported for hemibiotrophic pathosystems have generally been greater than 1.0 and often greater than 3.0. For example, Erickson et al. (2003) reported 6.1 for *Marssonina brunnea* f. sp. *brunnea* on poplar (*Populus* spp.), Bassanezi et al. (2001) reported 3.8 for *Phaeoisariopsis griseola* (Sacc.) Ferr. on common bean, and Roloff et al. (2004) found values of 2.8 and 3.1 for *Septoria albopunctata* Cooke on *Vaccinium* spp. Bourgeois and Boote (1992) found a reduction of 65% in the photosynthesis of peanut leaflets with 15% LLS disease (corresponds to a β value of around 4.0). Consequently, greater reductions in photosynthesis beyond the visual lesion area seem to be more common with hemibiotrophs compared to biotrophs.

Reasons for reduced photosynthesis beyond the measured visual lesion area, and thus differences in *β* are still not clear, but have been related to reductions in chlorophyll (Lopes and Berger, 2001; Moriondo et al., 2005) and carboxylation efficiency. Nogues et al. (2002) concluded that decreased maximum carboxylation velocity of Rubisco ($V_{c,max}$) was likely the primary determinant underlying the decline in photosynthetic rate of tomato (*Lycopersicon esculentum* Mill.) leaves infected by *Fusarium oxysporum* f.sp. *lycopersici*.

Although little difference in *β* values are generally observed among cultivars infected by biotrophic pathogens (Bassanezi et al., 2001; Kumudini et al., 2010),

genotypic differences in β have been observed among cultivars in response to hemibiotrophic pathogens (Erickson et al., 2003).

Simulations of Late Leaf Spot Damage

Crop-disease interactions have traditionally been quantified as damage functions (Pinnschmidt et al., 1994) consisting of empirical regression equations. However, these equations were very specific to a given condition and could not be used under different environmental conditions. Estimating the effects of disease epidemics on crop cultivars grown under different environmental and management conditions requires the use of mechanistic crop growth models (Teng et al., 1998). A generic approach whereby disease-induced damage is recorded and used as input to crop models has been successfully used by number of researchers to simulate effects of disease on growth and yield reduction of crops (Pinnschmidt et al., 1995; Teng et al., 1998; Batchelor et al., 1993; Boote et al., 1993). This approach can also be used to pinpoint the relative importance of the damage mechanisms and to identify gaps in knowledge of disease effects. The coupling points for damage mechanisms are located at the plant process level (photosynthesis, respiration, etc.) or at the state level (tissue area, weight etc.) (Boote et al.,1983a). Models that have been used to predict the impact of foliar diseases like LLS on growth and yield have generally incorporated the disease effects on defoliation and photosynthesis (Batchelor et al., 1993; Williams and Boote, 1995; Naab et al., 2004; Adomou et al., 2005).

The CROPGRO Model

The CROPGRO-Peanut model (Boote et al., 1998a, 1998b) is a process-oriented mechanistic crop growth model which considers crop carbon balance, crop and soil N balance, and soil water balance at the process level. This model has coupling points

and procedures for entering pest damage to simulate growth and yield reductions associated with foliar pathogens like LLS (Batchelor et al., 1993; Boote et al., 1993; Teng et al., 1998). The primary impacts of disease are simulated as defoliation; however the impacts of virtual lesion area are currently simulated by simply defoliating more leaf area (hence zero photosynthesis on that area) rather than creating direct impact at the leaf level photosynthesis (Adomou et al., 2005). This subroutine has been tested by some previous studies (Naab et al., 2004; Adomou et al., 2005) to simulate LLS effects on peanut growth and yield. However, these studies did not include measured data on leaf necrosis and defoliation required by the disease subroutine in the model. Either visual ratings were linearly regressed against measured necrosis range (minimum of 0% to maximum of 9%) from other studies (Bourgeois et al., 1991) to obtain necrosis values (Adomou et al., 2005) or variable defoliation and necrosis values were used to mimic leaf weight loss (Naab et al., 2004). This signifies the need of having reliable disease evaluation methods to provide accurate assessment of disease effects, which could then be used as input to crop growth models and as evaluation methods in breeding programs.

Model Evaluation

Statistics used for model evaluation vary from simply summing the difference between predicted and measured values to calculating more complicated concordance correlation coefficients (Lin et al., 2002). Two measures that are widely reported in the literature are the root mean squared error (RMSE) and the Willmott agreement index (Willmott 1981, 1982). The RMSE reflects the magnitude of the root mean sum of square differences between the predicted (P) and observed (O) values over time and is calculated as:

$$RMSE = \sqrt{\frac{\sum_{i=1}^{n}(P_i - O_i)^2}{n}})$$

The D-index is a descriptive index that measures dispersion of the simulated and observed data, calculated as:

$$D - index = 1 - \left[\frac{\sum_{i=1}^{n}(P_i - O_i)^2}{\sum_{i=1}^{n}\left(| P_i - \overline{O} | + | O_i - \overline{O} | \right)^2} \right]$$

Where n is the total number of observations, P_i is the predicted value for the ith measurement, O_i is the observed value for the ith measurement, and $\overline{O}$ is the overall mean of the observed values. A model performs well when the RMSE approaches zero and the D-index is close to 1.0.

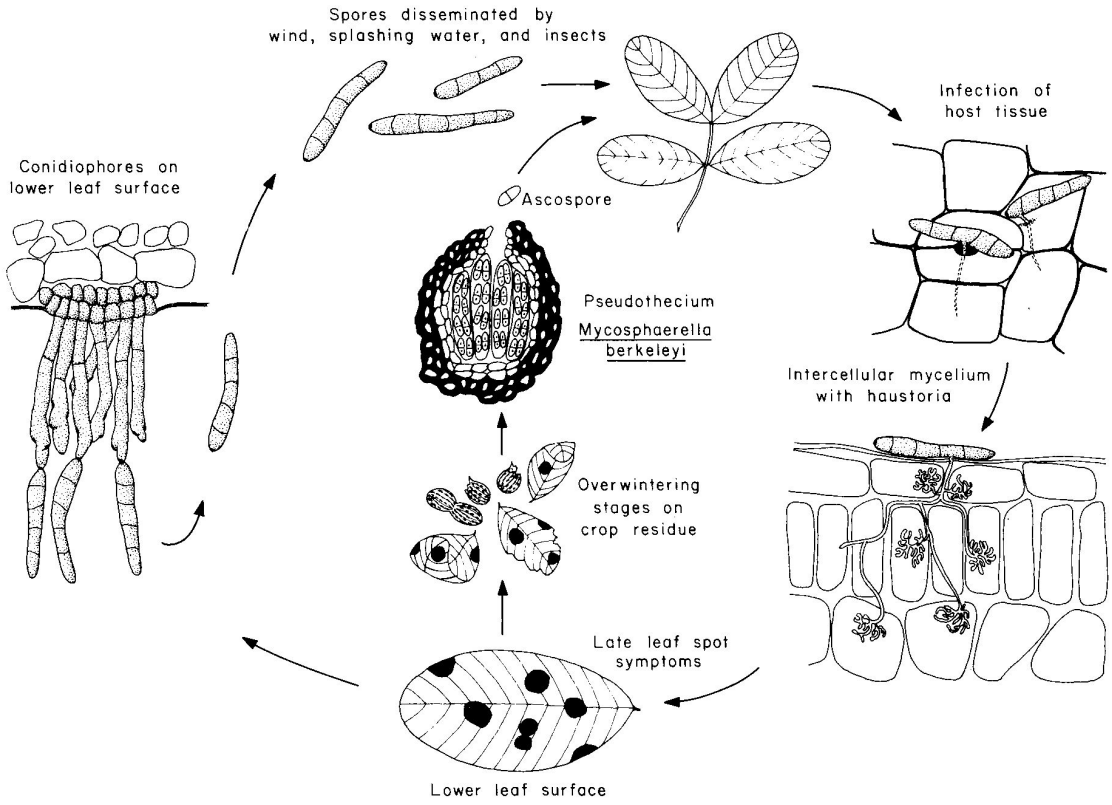

Figure 2-1. Disease cycle of late leaf spot, caused by Cercosporidium personatum (Berk. and Curt.) Deighton (reprinted with permission from Shokes and Culbreath, 1997).

CHAPTER 3
LATE LEAF SPOT EFFECTS ON GROWTH, PHOTOSYNTHESIS, AND YIELD IN PEANUT CULTIVARS OF DIFFERING RESISTANCE

Abstract

Cercosporidium personatum (Berk. & Curt.) Deighton causes late leaf spot (LLS) in peanut (*Arachis hypogaea* L.), which leads to necrotic lesions, early leaf senescence and yield losses. Detailed physiological analyses can contribute to an improved understanding of peanut-disease interactions and cultivar improvement. A study was conducted evaluating two peanut cultivars with more (York) and less (Carver) quantitative resistance to *C. personatum* grown under fungicide-sprayed and non-sprayed conditions in the field at Citra, Florida over two years. Data were collected on disease severity using the Florida 1 to 10 visual rating scale and by direct measurement of percent canopy lesion area. Leaf lifespan, total canopy photosynthesis (TCP), plant growth, and pod yield were also measured. Disease severity based on canopy lesion area was reduced by 30% in York compared to Carver. No additive effects of combining the resistant cultivar with fungicide were seen, as fungicide use increased yield by 364 kg ha^{-1} for both cultivars. Yield was more strongly related to disease severity based on canopy lesion area than to the Florida scale. Yield improvement with York was not as closely related to disease severity with only a 6% gain in pod yield in York compared to Carver. In addition, reduction in TCP was greater in York compared to Carver given their respective disease severity. These results indicated that combining resistance with the maintenance of physiological function during LLS infection could result in improved peanut yields under diseased conditions.

Background

Peanut (*Arachis hypogaea* L.) is one of the major sources of protein and oil in the world. It is cultivated on 24 million ha in more than 100 countries, generating an annual production of nearly 37 Tg (FAO, 2011). Nevertheless, worldwide peanut production is severely hampered by the incidence of numerous diseases. Early leaf spot (caused by *Cercospora arachidicola* S. Hori), and late leaf spot [caused by *Cercosporidium personatum* (Berk. & Curt.) Deighton] are among the most widespread and damaging foliar diseases of peanut in the southeastern United States (Nutter Jr. and Shokes, 1995). Pod yield losses can be greater than 50% when fungicides are not applied (Shokes and Culbreath, 1997). In Florida, late leaf spot (LLS) is the predominant disease (Jackson, 1981), causing yield losses of up to 50% (Pixley et al., 1990a). Consequently, regular and costly fungicide applications are currently used to minimize yield losses from peanut diseases (Woodward et al., 2008; Monfort et al., 2004). Improved cultivars with moderate resistance to late leaf spot, along with other integrated disease management practices, have also been successfully used to reduce inputs and production costs (Woodward et al., 2010; Woodward et al., 2008; Monfort et al., 2004). However, the effects of LLS on the physiological responses in cultivars of differing leaf spot resistance is not well understood and could contribute to improved cultivar development for disease resistance.

Cercosporidium personatum is a hemibiotrophic fungal pathogen that infects peanut leaves and stems (Mims et al., 1988). The initial source of inoculum is primarily conidia from crop residues in the soil. Conidia are rain-splashed or wind-blown onto leaf surfaces where they initiate infection. Symptoms are first recognizable as small necrotic flecks that enlarge to dark brown lesions from 1 to 10 mm in size (Smith et al., 1992).

Lesions generally develop within 10-14 days of initial infection. Symptoms are influenced by host genotype and environmental conditions, such as high temperature, rainfall, and relative humidity (Shew et al., 1988).

Peanut economic yield is a function of cumulative biomass and harvest index, which is determined by partitioning of assimilates to pod and effective duration of pod fill (Phakamas et al., 2008; Williams and Boote, 1995; Duncan et al., 1978). Premature loss of green leaf area (by necrotic tissue and defoliation) and reduction of leaf photosynthetic capacity due to disease contribute to a loss of canopy carbon assimilation, and thus a loss of yield. Many older peanut cultivars such as Florunner and Georgia Green have poor resistance to LLS. Loss of leaf area due to accelerated senescence was reported to be the predominant mechanism of yield loss in these cultivars (Bourgeois and Boote, 1992; Boote et al., 1980). However, for cultivars with improved resistance to LLS that experience less defoliation, yield reduction may also be related to leaf physiological response to disease instead of to loss of leaf area alone.

The breeding and selection of cultivars with partial resistance to LLS has been an important part of integrated disease management programs for reducing yield losses in peanut. Several new releases have shown good resistance associated with delayed disease progress and decreased defoliation. Components of resistance identified include extended latent period of the fungus, reduced sporulation and smaller lesion diameters (Chiteka et al., 1988; Dwivedi et al., 2002; Cantonwine et al., 2008). Selection of these resistant cultivars is typically based on visual disease ratings (e.g., Florida 1 to 10 scale) that combine both visual lesion disease severity and defoliation (Gorbet and Tillman, 2008). Direct measures of canopy lesion severity using image analysis may

improve estimates of disease severity, especially in resistant cultivars that exhibit decreased defoliation. While these measures of disease severity work well for monitoring disease dynamics, they do not always correlate well with yield reductions (Bergamin Filho et al., 1997; Jesus Jr. et al., 2001), due to a disconnect between the ratings and actual functional impairment (Bastiaans, 1991). In addition, host functional response to pathogens can be variable depending on environment, genotype, and physiological status (Zhang et al., 2009; Erickson et al., 2003).

Better understanding of the physiological responses to LLS related to yield in cultivars differing in resistance is needed to contribute to improved cultivar selection and modeling growth and yield responses of peanut to leaf spot. The objective of this study was to characterize LLS severity and its effects on growth and partitioning, leaf lifespan, canopy photosynthesis, and pod yield of York, a relatively resistant cultivar, compared to Carver, a cultivar with relatively poor resistance to LLS in a field environment.

Materials and Methods

Experimental Site and Design

Field experiments were conducted during the 2008 and 2009 growing seasons at the Plant Science Research and Education Unit in Citra, Florida (29^{o}23'60'' N, 82^{o}12'0'' W) on a Gainesville loamy sand (Hyperthermic, coated Typic Quartzipsamments) soil. The experiment was a multi-factorial design with the main factors being cultivar, fungicide application and year. Cultivar and fungicide application were arranged in a randomized complete block (RCB) with four replications of each treatment. Two cultivars were selected for differences in resistance to LLS: Carver (Gorbet, 2006) has poor resistance to LLS; while York (Gorbet and Tillman, 2011) has moderate resistance to LLS (Tillman et al., 2008). Fungicide application included: (i) no fungicide application;

and (ii) an industry standard fungicide schedule (Table 3-1) applied on a 14-day interval commencing from approximately 40 DAP. Fungicides were applied using a CO_2 backpack sprayer calibrated to deliver 328 and 374 L ha^{-1} during 2008 and 2009, respectively. A hand-held boom with five flat fan nozzles, spaced 45.7 cm apart was used to spray two rows at a time (spray coverage of 182 cm wide).

Plots were previously sown with bahiagrass (*Paspalum Notatum* Flueggé) and rye (*Secale cereale* L.) in a four year rotation with rye (nurse crop to establish bahiagrass) followed by two years of bahiagrass and then peanut. Sowing occurred during the latter part of the recommended planting window for North Central Florida on May 20 in 2008 and May 27 in 2009 to maximize LLS pressure (Wright et al., 2006). Each plot consisted of 6 rows spaced 0.91 m apart and 4.6 m long. Each block was separated by 3.7 m fallow alleys and the entire study was surrounded by two border rows. Seeds were sown at a rate of 17-20 seeds per meter row using a conventional planter. In-furrow application of azoxystrobin was conducted at a rate of 0.16 kg a.i. ha^{-1} while planting to control seedling diseases. Irrigation was applied as needed with a linear move system. Standard management practices for irrigated peanuts were employed during both years (Wright et al. 2006), including a 3-9-18 blended granular fertilizer that was broadcast before planting at a rate of 560 kg fertilizer ha^{-1} during both growing seasons. To satisfy the calcium requirement for pod and kernel formation, gypsum was broadcast at a rate of 2240 kg ha^{-1} split equally in two applications around 35-40 DAP followed by another application 10-14 days later. Disodium octaborate tetrahydrate was applied with the first two fungicide sprays at a rate of 5.6 kg ha^{-1} per application to supply boron.

Measures of Disease Severity and Growth

Late leaf spot intensity was assessed with the widely used Florida 1 to 10 scale (Table B-1, Woodward et al., 2010; Gorbet and Tillman, 2008; Cantonwine et al., 2008; Chiteka et al., 1988). Values of 1 to 4 indicate increasing leaf spot incidence on leaflets within the lower or upper canopy, but no defoliation. Ratings from 4 to 10 are associated with increasing levels of defoliation (Chiteka et al., 1988). Ratings began when visual symptoms first appeared (87 and 77 DAP in 2008 and 2009, respectively) and continued every 7-10 days until harvest. Area under disease progress curve (AUDPC) values were calculated for each plot from these disease ratings (Shanner and Finney, 1977) and were standardized by dividing AUDPC values by the number of days from the first observed symptoms to harvest to account for differences in the duration of LLS epidemics (Woodward et al., 2010; Woodward et al., 2008). Microscopic examination of lesions on leaflets indicated that *C. personatum* was the dominant pathogen in both years (Figure 3-1). Tomato spotted wilt (caused by Tomato spotted wilt virus) and white mold (caused by *Sclerotium rolfsii* Sacc.) was not observed in the field plots during both growing seasons.

Canopy defoliation and disease severity, the components that make up the Florida scale ratings, were also measured objectively throughout the growing season to compare to the more subjective Florida 1 to 10 scale assessment. Approximately biweekly, a randomly selected 61cm segment of the outer two rows of each plot was harvested, minimizing disturbance or border effects on the inner two final harvest yield rows. A representative subsample excluding the largest and the smallest plants was selected from each harvested sample (Bourgeois et al., 1991; Pixley et al., 1990a). Forty leaflets were randomly selected throughout the canopy from this subsample plant.

All leaflets were scanned at 300 dpi using a flatbed scanner (Microtek ScanMaker 5800, Microtek Int. Inc., Industrial Park Hsinchu, Taiwan) and stored as .tiff files. Leaf images were processed using ASSESS ver 2.0 image analysis software (American Phytopathological Society, St. Paul, MN) to give the percent canopy lesion area (Figure B-2, Erickson et al., 2003). AUDPC values for serial measurements of canopy lesion area were calculated and standardized for each plot similarly to the AUDPC values from the Florida 1 to 10 scale disease progression assessment. The remaining harvested sample was immediately oven dried for 72 h at 60^{o}C and subsequently weighed. Leaflets and pods were separated from all subsamples. Pods were counted and then leaves, stems and pods were oven dried to a constant mass. Stem, leaf and pod dry weights (DW) were determined for the entire sample by multiplying their respective fractions of the subsample times the total weight of the harvested sample.

In the central two rows of each 6-row plot, five plants were chosen at random and the first fully expanded leaf on each main stem was tagged using colored plastic tags at 49 and 92 DAP in 2008, and at 50, 65, and 79 DAP in 2009. These leaves were examined at weekly intervals until defoliation to calculate the total leaf lifespan in days for all the leaflets.

Measures of Canopy Photosynthesis and Yield

Starting approximately 35 DAP, a 61cm section of row was selected randomly from the outer two rows to measure canopy photosynthesis. Measurements were taken at 10-15 d intervals, using a 91 cm x 61 cm aluminum-frame mylar chamber and a portable photosynthesis system (LICOR LI-6200, Li-Cor Inc., Lincoln, NE) as explained by Bourgeois and Boote (1992). Carbon exchange rate was measured on two plots from each treatment under full sunlight and total darkness (achieved by covering the large

chamber with a black plastic sheet) conditions between 10:00 and 14:00 h. Measured carbon exchange rates under dark conditions were considered to represent canopy, root, and soil respiration. Total canopy photosynthesis (TCP, Boote et al., 1983b) was calculated by adding the absolute dark respiration to the observed carbon exchange rate.

The central two rows of each 6-row plot in each genotype were dug at maturity (determined by hull scrape method; Williams and Drexler, 1981) using a conventional two-row digger-shaker-inverter. Plants were allowed to sun-dry in the field for 3-4 days. Afterwards, stationary threshers were used to harvest pods. Peanut yields were determined after drying to uniform moisture content of 9% (wt/wt). Sprayed plots of Carver were inverted 135 and 127 DAP in 2008 and 2009, respectively. Non-sprayed plots were harvested approx. 7 days earlier in each year due to leaf spot pressure. Both sprayed and non-sprayed York plots were inverted 149 and 145 DAP, respectively.

In 2009, a subsample of 200 g of pods per plot was subjected to a standard analysis for peanut grade. Pod samples were graded using standard farmer stock grading equipment in accordance with the federal-state inspection service method. Pod grades were defined as percent total sound mature kernels (TSMK) which is the sum of sound mature kernels and sound split kernels.

Statistical Analysis

Statistical analyses were performed using analysis of variance procedures in the GLIMMIX procedure of SAS (SAS Institute, 2009). Cultivar, fungicide regime, year, and their interactions were considered fixed effects and block by year as a random effect. Degrees of freedom were determined using the Kenward-Roger method. Where significant ($P < 0.05$) fixed effects were seen, pairwise comparisons were made using

the LSMEANS statement with TUKEY method. Relations between yield and disease severity were analyzed using linear regression procedures.

Statistical analyses of total biomass and its partitioning, and TCP were performed using nonlinear regression procedures of the nlme library of R (R Development Core Team, 2008). A 3-parameter logistic function (Eq. [3] in Yin et al., 2003) was employed to fit stem, pod, and total biomass data, which provided a y-asymptote value, shape parameter related to growth rate, and DAP value at inflection point, which represents the DAP at half of the maximum value on the y-axis. Leaf weight and TCP were fit with a 3-parameter gaussian function (Gauch Jr. and Chase, 1974), which provided the maximum value on the y-axis, DAP at which the maximum value was achieved, and a peak width parameter at ½ of the maximum value. Analysis of variance was run on these parameters using GLIMMIX of SAS, as explained earlier (except for TCP as data was collected for only two replicates). Results of this analysis are reported only when significant.

Results

Growth Environment

Environmental conditions during the 2008 and 2009 growing seasons were quite favorable for LLS development (Figure 3-2). Rainfall from mid-May through harvest in mid-October was 481 and 745 mm in 2008 and 2009, respectively. This precipitation was received in 58 events in 2008 and 74 events in 2009. Irrigation was not applied in either year after onset of disease as rainfall was adequate for crop growth. Average daily temperature during the same period was 25.8 and 25.5ºC in 2008 and 2009 respectively. Relative Humidity ranged from 62 to 96% in 2008 and 65 to 96% in 2009.

Disease Assessment

Late leaf spot epidemics occurred in both years of the study, but appeared earlier in 2009 compared to 2008 (Figure 3-3) consistent with more frequent and abundant rainfall in 2009 compared to 2008. Late leaf spot symptoms were first observed visually in the field around 95 and 80 DAP during 2008 and 2009 on both cultivars, respectively. Carver, the less resistant cultivar, showed more rapid disease progress than York during both years, especially in non-sprayed plots. Fungicide delayed the initial progress of disease symptoms in both cultivars (Figure 3-3).

Standardized values for the area under disease progress curves for both Florida 1 to 10 scale ratings ($stAUDPC_{FL}$) and percent canopy necrotic lesion area ($stAUDPC_{Les}$) were generally in good agreement and showed significantly reduced disease intensity associated with fungicide inputs and with the moderately resistant cultivar York compared to the poorly resistant cultivar Carver (Table 3-2). For example, $stAUDPC_{Les}$ and $stAUDPC_{FL}$ were 30% and 19% lower in York compared to Carver, respectively. Similarly, fungicide-sprayed plots showed a 43% reduction in $stAUDPC_{Les}$ and a 26% reduction in $stAUDPC_{FL}$ compared to non-sprayed plots. A significant year x cultivar x fungicide effect on $stAUDPC_{Les}$ resulted from higher values in York in 2008 compared to 2009, whereas higher values in Carver were seen in 2009 compared to 2008 (Table 3-2). This pattern was not seen in $stAUDPC_{FL}$, as 2009 values were significantly higher in both cultivars, resulting in a significant year effect.

Plant Growth and Development

Although the cultivars did not differ ($P > 0.05$) in their maximum stem or leaf DW (or leaf area index, data not shown), Carver achieved maximum leaf DW 10 days earlier ($P = 0.03$, Figure 3-4) and the DAP value at inflection was 10 days earlier ($P < 0.001$)

for stem DW. Maximum leaf DW was attained at 79 and 89 DAP in Carver and York, respectively, across both growing seasons. In both years, following attainment of maximum leaf DW, defoliation was observed in all treatments, but defoliation in non-sprayed plots generally exceeded that of fungicide-sprayed plots, as indicated by narrower peak widths for leaf DW ($P < 0.01$). This effect was greater in Carver compared to York as leaf lifespan data of tagged leaf cohorts showed greater differences in leaf lifespan in sprayed plots compared to non-sprayed plots for Carver (Table 3-3). In addition, defoliation occurred more quickly and to a greater extent in Carver compared to York, as indicated by narrower peak widths ($P = 0.03$) in Carver (Figure 3-4). Notably, partitioning to leaf and stem weight largely occurred before appreciable disease was found, whereas much of the partitioning to pod weight occurred after disease (Figure 3-4). For example, DAP at inflection for stem weight in Carver was at 53, while DAP for pod weight was 82. In addition, DAP value at inflection for pod weight occurred sooner ($P < 0.001$) in Carver (82 DAP) compared to York (101 DAP).

Canopy photosynthesis was in agreement with seasonal patterns of leaf and stem accumulation, as maximum TCP occurred at 70 DAP in Carver and 80 DAP in York, but maximum TCP was similar between cultivars (Figure 3-5). In addition, similar peak width values indicated similar declines in TCP between cultivars, despite disease progress that was comparatively slower in York than Carver (Figure 3-3). However, fungicide application resulted in a slower decline in TCP, as indicated by a peak width of 34 days in fungicide-sprayed plots compared to 28 days in their non-sprayed counterparts (Figure 3-5).

Pod Yield and Quality

Mean pod yields across all treatments ranged from 2500 to 3500 kg ha^{-1} (Table 3-2). Fungicide application resulted in a significant increase in pod yield (12.5%) over non-sprayed plots. However, there was a significant year x fungicide interaction whereby differences were significant in 2009, but not in 2008. Averaged across all treatments, pod yields were not different among growing seasons (P = 0.71). This was due to a significant cultivar x year interaction, whereby the poorly resistant cultivar (Carver) outyielded the moderately resistant cultivar (York) in 2008, whereas the opposite was true in 2009. Notably, there was no cultivar x fungicide interaction seen in either year of the study, indicating no diminished response of fungicide on absolute yield gain of York. Averaged across all treatments, number of pods per unit area was greater (P = 0.03) while average pod size was smaller (P < 0.01) in 2009 compared to 2008. Pod yield was negatively related to $stAUDPC_{FL}$ and $stAUDPC_{Les}$ and the slopes of these relationships were not affected by cultivar or fungicide schedule (Figure 3-6). Overall, the relationship between pod yield and $stAUDPC_{Les}$ was better than that between pod yield and $stAUDPC_{FL}$, which was especially evident at relatively low disease severities. Finally, neither cultivar nor fungicide affected peanut TSMK during 2009 (Table 3-2).

Discussion

The overall objective of this study was to gain an improved understanding of peanut response to disease by looking at effects of LLS on peanut physiology, growth and yield of two cultivars differing in resistance, which will be important for continued cultivar improvement and lower fungicide input in peanut production. The more resistant cultivar contributed to delayed disease progress, which resulted in slower development of canopy lesion area and less defoliation. Improved yield in the more resistant cultivar

was seen in one year of the study when the LLS disease severity was high. No additive effects of combining improved cultivar resistance and application of fungicide on pod yield were found, as the absolute gains in yield associated with fungicide treatment were the same between both cultivars across both years of the study. Pod yield was better related to $stAUDPC_{Les}$ compared to $stAUDPC_{FL}$. Finally, TCP was found to decline similarly in both cultivars despite the slower progress of disease noted in the more resistant cultivar.

Delayed disease progress in more resistant cultivars like that seen in the present study has been demonstrated in other studies using the Florida 1-10 scale ratings (Woodward et al., 2010; Monfort et al., 2004) and canopy disease severity (Pixley et al., 1990b). Visual disease presence in the improved cultivar appeared to start at the same time as in the less resistant cultivar during both years; however, the progress of the disease was slower in the improved cultivar. This differing pattern of disease progress could be explained by a number of factors including a reduced number of initial infection points (foci) and/or differences in the latent period of the fungus. Prior studies have found little difference in the incubation period among a wide range of peanut genotypes, while the latent period tended to be longer in more resistant genotypes, resulting in slower temporal progression of the disease (Cantonwine et al., 2008; Dwivedi et al., 2002; Chiteka et al., 1988).

Although the 14-day calendar-based fungicide program did not achieve 100% disease control, fungicide application delayed the progress of disease symptoms (Pixley et al., 1990b; Bourgeois et al., 1991). Substantial necrosis and defoliation due to LLS was observed in the control plots (Figures 3-3, 3-4 and 3-5; Table 3-3) during both

growing seasons which is typical for non-sprayed peanut. This was also observed by other studies conducted under different growing seasons and locations (Woodward et al., 2010; Monfort et al., 2004). This study also showed that yield benefits associated with applying fungicide did not differ significantly between cultivars varying in their resistance to LLS. Therefore, based on the results, growers might be reluctant to reduce fungicide applications even on more resistant cultivars. However, other studies have shown non-significant yield losses in more resistant cultivars with reduced fungicide application compared to a 14-day calendar-based schedule (Woodward et al., 2010; Monfort et al., 2004). This discrepancy might be due to differences in peanut cultivars, LLS severity, environment and/or fungicide schedule.

Resistance to LLS in southeastern U.S. runner-type peanut cultivars has generally been associated with later maturing varieties that possess a later onset of pod fill and a reduced pod growth rate, but possess longer effective pod fill duration (Pixley et al., 1990a). In the present study, York showed later initiation of pod fill, slower pod growth rate, and longer duration of pod fill compared to Carver (Figure 3-4). Implications of these growth patterns for LLS effects on yield depended on onset of the disease epidemic in this study. In 2008, when LLS was relatively late in arrival, partitioning to pod yield was nearly complete in Carver, and thus relatively high yields were attained with Carver with little effect of fungicide on yield. In contrast, in 2009 when LLS arrived about 2 weeks earlier compared to 2008, LLS effects on pod yield were greater and effect of fungicide was greater. Thus, where later planting dates are desired (e.g., to minimize incidence of tomato spotted wilt virus), cultivars with improved LLS resistance are beneficial. Finally, since LLS had no effect on TSMK or average pod size in this

study (Table 3-2), the determinant of yield impacted by LLS was pod number, which is consistent with the Phakamas et al. (2008) study that showed that peanut yield was primarily determined by pod number and not pod size across genotypes.

Relations between yield and disease severity measurements are often weak (Jesus Jr. et al., 2001); however significant regression relationships were found in this study (Figure 3-6). This finding may be due to the wide ranges of disease severity and yield observed in this study. In addition, yield was more strongly related to $stAUDPC_{Les}$ compared to $stAUDPC_{FL}$. This suggests that pod yield response to disease epidemics is better explained by measured canopy lesion area rather than the visually determined Florida 1 to 10 scale, which is likely due to the fact that $stAUDPC_{Les}$ was determined using an objective digital image analysis instead of subjective visual ratings.

While pod yield reductions were generally related to disease ratings (Figure 3-6), yield reductions in York due to LLS were greater than the ratings indicated. Reduction in disease severity under non-sprayed conditions in York compared to Carver were 22 and 34% based on $stAUDPC_{FL}$ and $stAUDPC_{Les}$, respectively. Moreover, the leaf lifespan in non-sprayed York was longer than Carver (Table 3-3). However, this relatively lower disease severity resulted in only 8% yield improvement in York compared to Carver. Thus, the yield improvement in York was not proportional to the reduction in disease severity in this study. One potential explanation for this disconnect between disease reduction and yield improvement is the existence of at least two separate mechanisms: (i) the ability to sustain leaf photosynthesis during disease progression; and (ii) resistance to the progression of disease symptoms. In this study, the more resistant cultivar, York, may lack the ability to sustain photosynthesis at a given disease severity.

This idea is supported by similar reductions in TCP in both York and Carver despite reduced disease severity in York (Figure 3-3). Thus, a combination of LLS resistance (i.e., delayed disease progress) combined with host physiological tolerance (i.e., maintenance of physiological function in the presence of disease) may offer the most promising approach for peanut cultivar improvement and reduced fungicide input production systems.

In conclusion, this study demonstrates that cultivar resistance is an important component for integrated disease management of LLS in peanut, particularly during years with high disease pressure. Nevertheless, no diminished effect of fungicide with improved cultivar on absolute yield gain was observed. So, foliar application of fungicide still seems to play an important role in minimizing damage caused by LLS epidemics. Despite substantial reduction in disease severity and defoliation in the resistant cultivar York, yield improvement over the less resistant cultivar, Carver, was marginal and most beneficial under heavy LLS pressure. These findings were attributed in part to a lack of improved physiological tolerance to LLS in York. These results indicate that combining resistance to disease progression with enhanced ability to sustain canopy photosynthetic capacity in the cultivar selection procedure could provide significant improvement in our efforts to improve peanut yields under diseased conditions.

Table 3-1. Fungicide spray schedule for the field experiments at Citra, FL.

Spray	Fungicide
1	Chorothalonil (1.26)†
2	Chorothalonil (1.26)
3	Pyraclostrobin (0.18)
4	Azoxystrobin (0.33)
5	Chorothalonil (0.63) + Tebuconazole (0.23)
6	Chorothalonil (0.63) + Tebuconazole (0.23)
7	Chorothalonil (1.26)
8	Chorothalonil (1.26)

† Numbers in the parentheses denote the rate of fungicide application (kg a.i. ha^{-1})

Table 3-2. Treatment means (*n* = 4) and analysis of variance results for standardized area under the disease progress curve for Florida 1 to 10 scale ($stAUDPC_{FL}$) and percent canopy lesion area ($stAUDPC_{Les}$), pod yield, pod number, average pod weight, and total sound mature kernels (TSMK). Fungicide treatments were no fungicide application (NF) and standard 14-day calendar based application (F). Cultivars were Carver (C) and York (Y).

Year	Cultivar	Fungicide	$stAUDPC_{FL}$	$stAUDPC_{Les}$	Pod Yield	Pod No.	Pod Weight	TSMK
					Kg ha^{-1}	m^{-2}	g	%
2008	C	NF	4.13	3.36	3098	406	0.95	-†
		F	3.11	2.23	3290	395	0.95	-
	Y	NF	3.45	2.97	2925	284	1.03	-
		F	2.71	2.06	3122	354	1.08	-
2009	C	NF	5.17	4.93	2498	509	0.90	72.7
		F	3.51	2.25	3144	533	0.92	75.2
	Y	NF	3.78	2.53	3136	511	0.87	74.9
		F	2.91	1.39	3556	550	0.90	74.7
SIGNIFICANCE								
Cultivar			***	***	*	ns	ns	ns
Fungicide			***	***	***	ns	ns	ns
Cult x Fung			***	**	ns	ns	ns	ns
Year			*	ns	ns	*	*	-
Cult x Year			**	***	***	*	*	-
Fung x Year			*	**	*	ns	ns	-
Cult x Fung x Year			ns	*	ns	ns	ns	-

***, **, * and ns = $P < 0.001$, $P < 0.01$, $P < 0.05$ and $P > 0.05$ respectively; † Data not recorded

Table 3-3. Treatment means (n = 4) for leaf lifespan of leaf cohorts tagged at different times (DAP) throughout the growing season. Fungicide treatments were no fungicide application (NF) and standard 14-day calendar based application (F). Cultivars were Carver (C) and York (Y).

		Tagging date (DAP)				
		2008		2009		
Cultivar	Fungicide	49	92	50	65	79
		d	d	d	d	d
C	NF	66ab†	29c	53b	41c	31c
	F	69a	38b	62a	50a	42a
Y	NF	63bc	44a	52b	43bc	35b
	F	60c	47a	47c	44b	42a

† Numbers followed by the same letter within a column do not differ (P > 0.05)

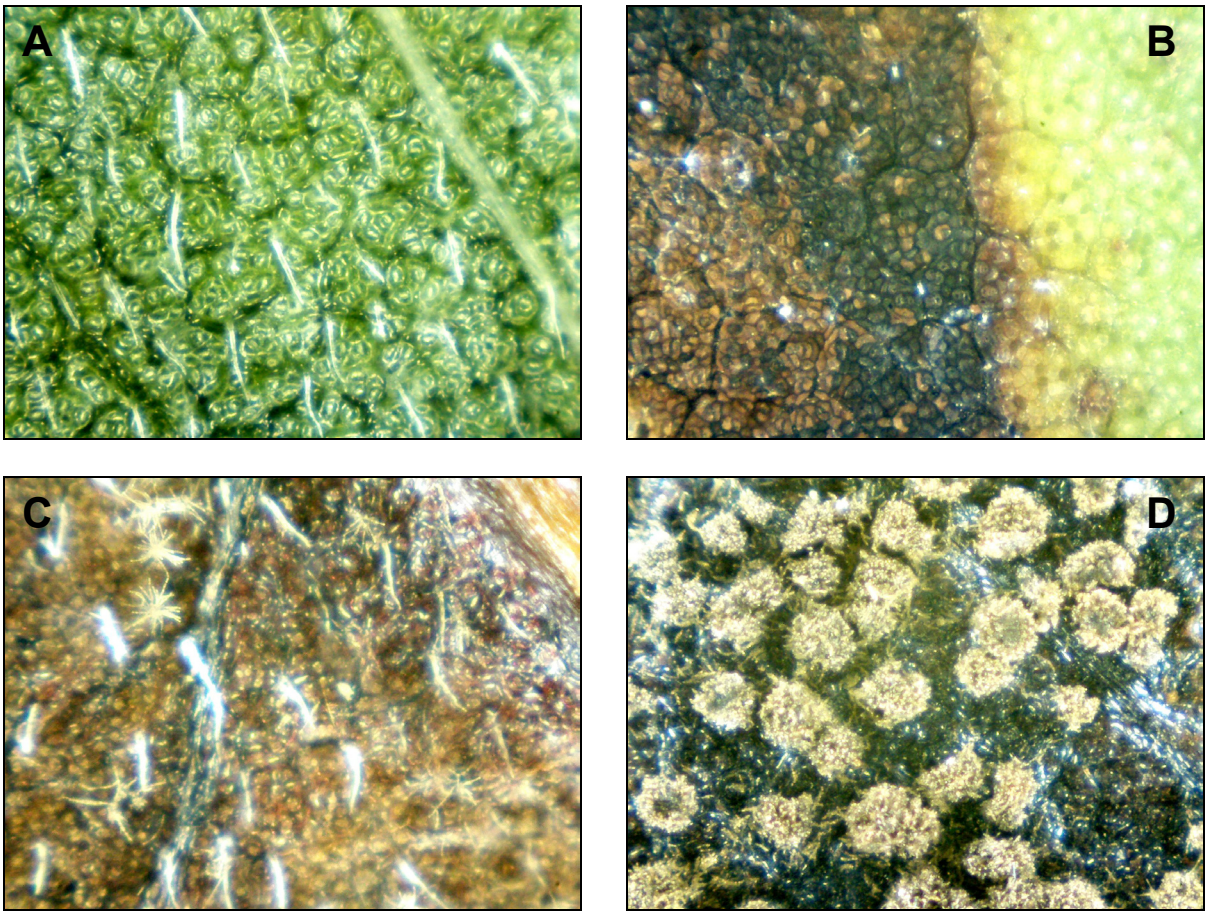

Figure 3-1. Peanut leaf with leaf spot disease. (A) abaxial side covered with fungal hyphae, (B) adaxial side with necrotic lesion but no conidiophores, (C) abaxial side with conidiophores of *Cercospora arachidicola*, and (D) abaxial side with conidiophores of *Cercosporidium personatum*.

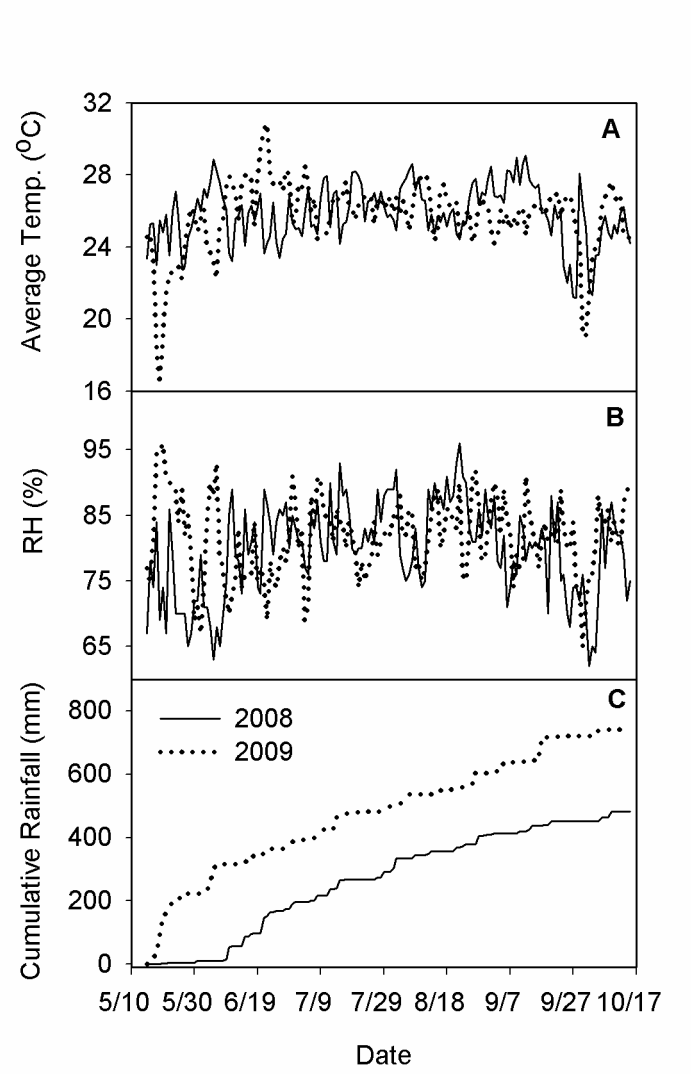

Figure 3-2. Average daily temperature (A), relative humidity (B), and cumulative rainfall (C) for the field experiment during the study period (Source: Florida Automated Weather Network, Citra, FL).

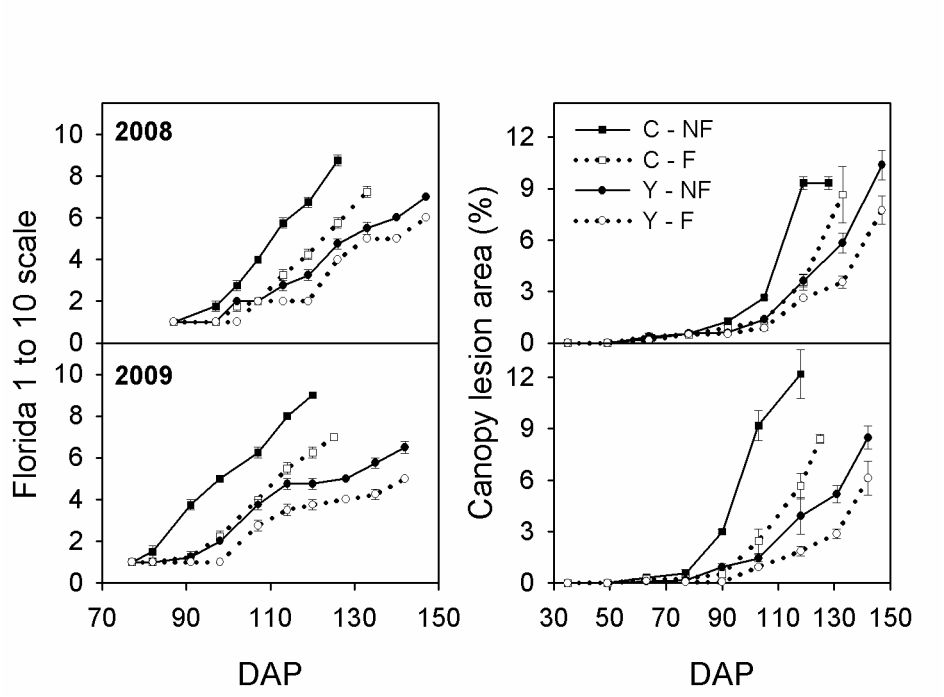

Figure 3-3. Progress of late leaf spot as estimated with the Florida 1 to10 scale and percent canopy lesion area during 2008 and 2009 growing seasons for the two peanut cultivars (C – Carver; Y – York) grown under fungicide sprayed (F) and non-sprayed (NF) conditions. Vertical bars greater than symbols represent ± standard error of the mean (*n*=4).

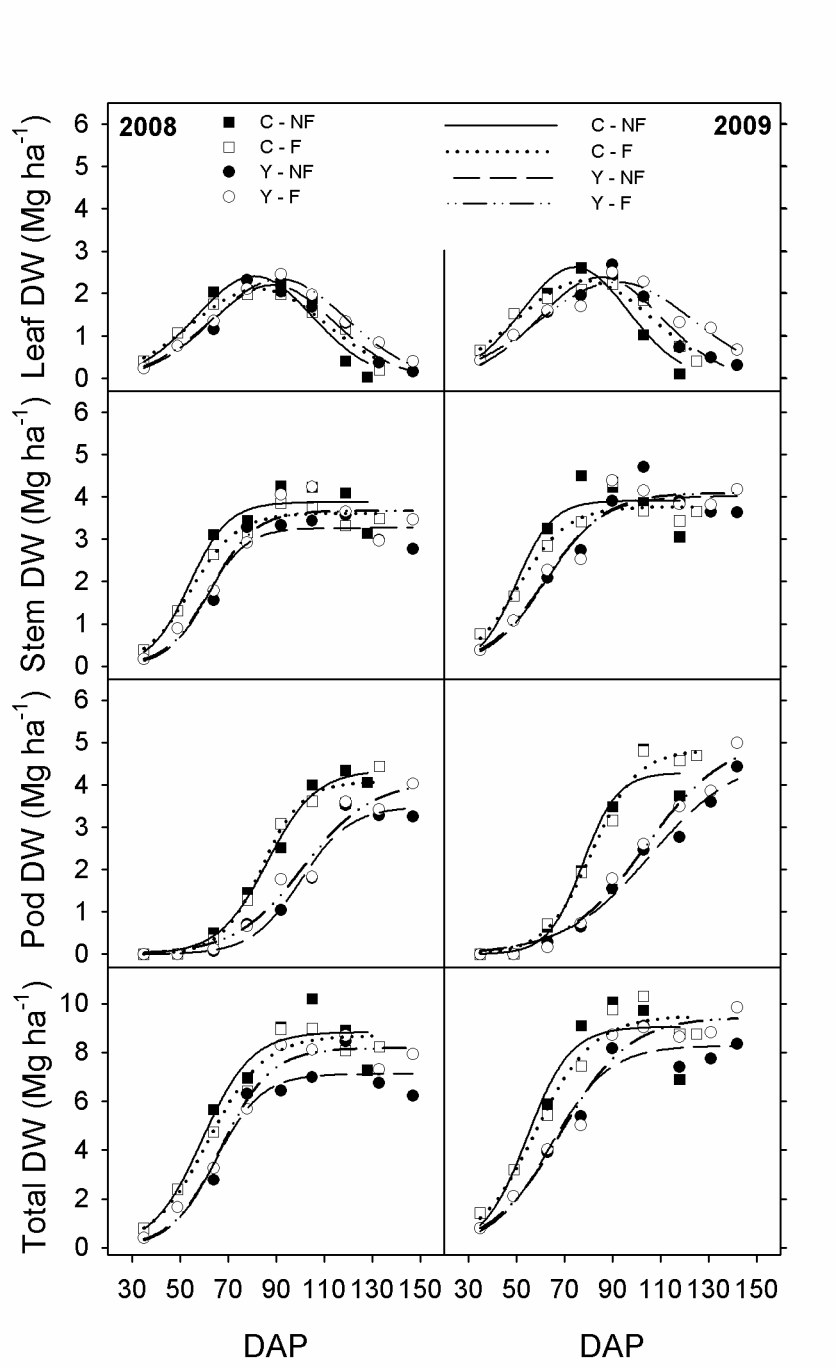

Figure 3-4. Leaf, stem, pod, and total dry matter accumulation vs. days after planting (DAP) for two peanut cultivars Carver (C) and York (Y) grown under fungicide sprayed (F) and non-sprayed (NF) conditions at Citra, FL during 2008 and 2009. Symbols represent treatment means (*n*=4) while regression lines represents gaussian (for leaf biomass) and logistic (for stem, pod and total biomass) model fits.

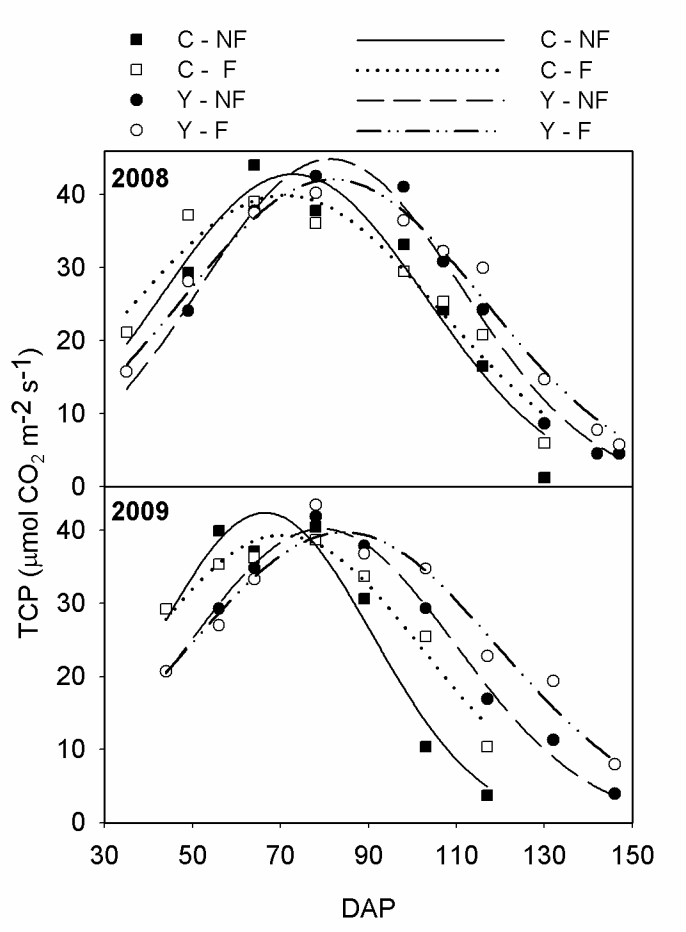

Figure 3-5. Mid-day total canopy photosynthesis (TCP) for two peanut cultivars (C - Carver; Y - York) grown under fungicide sprayed (F) and non-sprayed (NF) conditions at Citra, FL during 2008 and 2009. Symbols represent treatment means (*n*=2) while regression lines represents gaussian model fits.

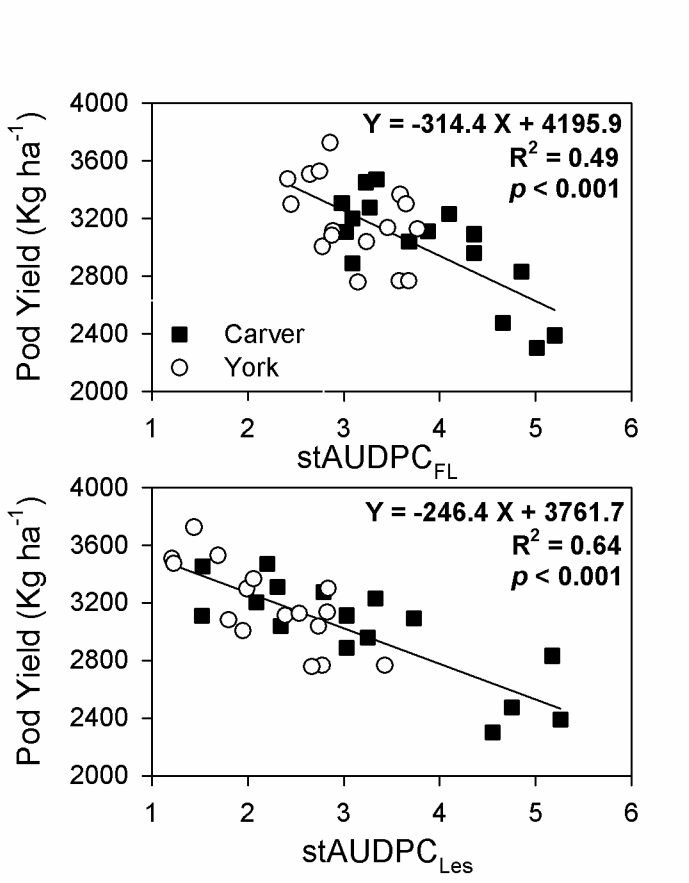

Figure 3-6. The relationship between pod yield and the standardized area under the disease progress curve based on the Florida 1 to 10 scale ($stAUDPC_{FL}$) and percent canopy lesion area ($stAUDPC_{Les}$) for the two peanut cultivars, Carver (C) and York (Y).

CHAPTER 4
PHOTOSYNTHETIC CONSEQUENCES OF LATE LEAF SPOT DIFFER BETWEEN TWO PEANUT CULTIVARS WITH VARIABLE LEVELS OF RESISTANCE

Abstract

Late leaf spot (LLS) caused by *Cercosporidium personatum* (Berk. & Curt.) Deighton reduces leaf CO_2 assimilation rate (A_{sat}) and accelerates leaf defoliation, which together lead to major reductions in peanut (*Arachis hypogaea* L.) yield worldwide. This study was conducted to determine whether differences in photosynthetic response to LLS severity exist among peanut cultivars of differing resistance. Field experiments were conducted in 2008 and 2009 to study the effects of LLS on A_{sat} of tagged leaf cohorts, and photosynthetic response of similar age leaves to LLS in peanut cultivars with more (York) and less (Carver) quantitative resistance. A non-linear model, $y = (1-x)^{\beta}$ was used to analyze A_{sat} data, where y is relative A_{sat}, x is measured visual lesion area, and β represents the relationship between virtual and visual lesion area. Progression of LLS severity on leaf cohorts was slower in York compared to Carver. However, the reduction in A_{sat} with leaf cohort age was similar across the cultivars. This paradox could be explained by a higher β value in York (4.6) compared to Carver (3.6), indicating a greater relative reduction in A_{sat} beyond the necrotic lesion area in York. This greater reduction in A_{sat} in York compared to Carver was most closely related to a reduction in maximum carboxylation velocity. Results indicated that future efforts to improve LLS resistance should include sustaining A_{sat} in response to LLS infection along with slower disease progress.

Background

Late leaf spot (LLS), caused by *Cercosporidium personatum* (Berk. & Curt.) Deighton, (telemorph = *Mycosphaerella berkeleyi* Jenk.) is among the most widespread

and damaging foliar diseases (Nutter Jr. and Shokes, 1995) of peanut (*Arachis hypogaea* L.) in the southeastern United States. Pod yield losses of up to 50% have been reported when fungicides are not applied (Shokes and Culbreath, 1997). Consequently, regular and costly fungicide applications are currently used to minimize yield losses from LLS (Monfort et al., 2004; Woodward et al., 2008). In addition, breeding and selection for improved cultivars with moderate resistance to leaf spot have recently been used with integrated disease management practices to reduce inputs and production costs (Monfort et al., 2004; Woodward et al., 2008). However, LLS effects on peanut yield are often variable and do not always correlate with cultivar LLS resistance ratings (Chapter 3). An improved understanding of the effects of LLS on leaf-level physiological responses could help to explain these variable yield responses and result in better identification of improved cultivars in breeding programs and better modeling estimates of peanut yield loss to LLS.

Cercosporidium personatum is a hemibiotrophic fungal pathogen that infects leaves, the photosynthetic unit of the peanut plant (Mims et al., 1988). Conidia, produced by conidiophores, on peanut residues in soil and off-season plants typically serve as a source of initial inoculum. Conidia are dispersed by wind, splashing water, and insects. Pathogenesis of *C. personatum* starts with the development of spore germination tubes which enter plant cells via stomata or directly through the epidermis, allowing intercellular mycelium growth. Lesions generally develop within 10-14 days of initial infection (Shokes and Culbreath, 1997) and cause reductions in leaf carbon assimilation, premature leaf senescence, and pod detachment that result in yield losses (Bourgeois et al., 1991; Bourgeois and Boote, 1992).

Several new releases from breeding programs (Branch, 2002; Holbrook and Culbreath, 2008; Tillman et al., 2008) have shown moderate resistance levels associated with slower disease progress and less disease-induced defoliation. Components of identified resistance include a lengthened latent period of the fungus, reduced sporulation, and smaller lesion diameters (Chiteka et al., 1988). Selection of resistant cultivars is typically based on visual disease ratings (e.g., Florida 1 to 10 scale) that combine both visual lesion disease severity and defoliation (Chiteka et al., 1988). While these ratings work well for monitoring disease dynamics, they do not always correlate well with yield reductions, especially at low disease severities (e.g., Bergamin Filho et al., 1997; Jesus Jr. et al., 2001). For example, similar yield losses between cultivars differing significantly in disease progress have been reported (Chapter 3). It has often been found that the lack of a correlation between disease severity and yield loss is due to a disconnect between disease ratings and the actual functional impairment, which has been related to a loss of photosynthetic activity beyond the visual lesion area (Bastiaans, 1991).

Reductions in the photosynthetic capacity of infected leaves have been shown in several pathosystems (Boote et al., 1983a; Bourgeois and Boote, 1992; Shtienberg, 1992; Kumudini et al., 2008). In order to relate reductions in leaf photosynthesis to visual lesion area, Bastiaans (1991) proposed a relatively simple model, $y = (1- x)^{\beta}$, where y is the relative net assimilation rate of a diseased leaf compared to that of an asymptomatic leaf, x is the measured visual lesion area, and β describes the relationship between virtual and visual lesion area. The virtual area represents loss of photosynthetic capacity beyond the visual lesion area. Thus, β indicates whether the

effect of disease on photosynthesis is higher ($\beta > 1$), lower ($\beta < 1$), or equal ($\beta = 1$) to that accounted for by the measured visual lesion area.

Using this model, several studies have shown that the relationship between photosynthesis and visual disease severity is related to host-pathogen interactions (Bassanezi et al., 2002; Erickson et al., 2003; Zhang et al., 2009). In general, studies have indicated that β values are relatively low (< 2.5) for biotrophic pathogens (Bassanezi et al., 2001; Lopes and Berger, 2001; Robert et al., 2005; Kumudini et al., 2010), intermediate (3 to 6) for hemibiotropic pathogens (Bassanezi et al., 2001; Erickson et al., 2003; Roloff et al., 2004) and highest for necrotrophic pathogens (Bassanezi et al., 2001; Lopes and Berger, 2001). Reasons for reduced photosynthesis beyond the measured visual lesion area, and thus differences in β are still not clear, but have been related to reductions in carboxylation efficiency (Nogues et al., 2002) and chlorophyll (Lopes and Berger, 2001; Moriondo et al., 2005). However, understanding why β values differ, and perhaps even more importantly, whether they differ by cultivar within species is critical for improved cultivar selection and for modeling effects of disease on carbon assimilation, growth and yield (Bastiaans, 1993; Adomou et al., 2005; Bancal et al., 2007). Although little difference in β values are generally observed among cultivars infected by biotrophic pathogens (Bassanezi et al., 2001; Kumudini et al., 2010), genotypic differences in β have been observed among cultivars in response to hemibiotrophic pathogens (Erickson et al., 2003).

Despite the importance of LLS in peanut production, there are no published β values for comparisons among peanut cultivars. The main objective of this study was therefore to evaluate and compare the effects of LLS severity on photosynthetic

metabolism in two peanut cultivars with variable levels of resistance to LLS. It was hypothesized that peanut cultivars would differ in their photosynthetic response to LLS, which could help explain variable yield losses due to disease (e.g., Chapter 3) and improve screening of cultivars and modeling growth and yield responses of peanut to LLS.

Material and Methods

Experimental Site and Design

Peanut leaves were sampled from field experiments conducted during the 2008 and 2009 growing seasons at the Plant Science Research and Education Unit in Citra, Florida (29°23'60'' N, 82°12'0'' W) to study leaf photosynthetic responses to LLS. These experiments were part of a larger study conducted to quantify the growth and yield losses due to LLS in peanut cultivars with variable levels of resistance (Chapter 3). The experiment was a 2x2 factorial arranged in a randomized complete block design with four replications. Cultivar and fungicide application were treated as fixed effects. . Two cultivars were selected for differences in resistance to LLS: Carver (Gorbet, 2006) has poor resistance to LLS; while York (Gorbet and Tillman, 2011) has moderate resistance to LLS (Tillman et al., 2008). Fungicide treatments included no fungicide application and an industry-standard fungicide schedule (Table 3-1 of Chapter 3) applied on a 14-d interval commencing from approximately 40 days after planting (DAP).

Plots were planted on 20 May in 2008 and 27 May in 2009 to maximize LLS pressure (Wright et al., 2006). Each plot consisted of six rows with a row spacing of 0.91 m and a row length of 4.6 m. Seeds were sown at a rate of 17 to 20 seeds per m of row using a conventional planter. In-furrow application of azoxystrobin was applied at a rate of 0.16 kg a.i. ha^{-1} to help control soilborne diseases. Irrigation was applied as

needed with an overhead linear move irrigation system. Plots were fertilized pre-plant with a 3-9-18 blended granular fertilizer at a rate of 560 kg fertilizer ha^{-1}. Gypsum was broadcast at a rate of 2240 kg ha^{-1} in two equal applications around 35 to 40 DAP followed by the second application 10 to 14 d later. Disodium octaborate tetrahydrate was applied with the first two sprays at a rate of 5.6 kg ha^{-1} to supply boron. A preemergence broadcast application of pendimethalin (0.92 kg a.i. ha-1) + diclosulam (0.42 kg a.i. ha^{-1}) and a postemergence application of imazapic (0.07 kg a.i. ha^{-1}) were used for weed control.

Measures of A_{sat} on Tagged Leaf Cohorts over Time

In the central two rows of each plot, five plants were chosen at random and most recent fully expanded leaves on each main stem were tagged using colored plastic tags at 49 and 92 days after planting (DAP) in 2008, and at 50, 65, and 79 DAP in 2009, respectively. At approximately weekly intervals, around five leaflets per cultivar were randomly selected and analyzed for A_{sat} on 6 cm^2 leaf area using a LI-6400XT portable, open-flow photosynthesis system (LI-COR Inc., Lincoln, NB). Measurements were made between 1000 to 1400 h under partly cloudy to cloud-free days at 1800 µmol m^{-2} s^{-1} photosynthetic photon flux density (PPFD) using the 6400-02 LED light source (LI-COR). The sample chamber CO_2 concentration was maintained at 400 µmol CO_2 mol^{-1} air, and the flow rate of air through the sample chamber was set at 500 µmol s^{-1}. Temperature was maintained at 30 ± 1°C and relative humidity was maintained between 60-70%, similar to environmental conditions.

Severity of LLS was assessed on all of the tagged leaflets at approximately weekly intervals until defoliation using the ICRISAT diagrammatic scale (Figure B-1, Subrahmanyam et al., 1995) to estimate the percent disease severity. The ICRISAT

scale is a visual diagrammatic scale which depicts the proportion of the leaf area (%) that has necrotic lesions. Disease severity was defined as the proportion of total necrotic lesion area to total leaf area. Microscopic examination of conidiophores and conidia on diseased leaves indicated that *C*. *personatum* was the dominant pathogen in both years (Figure 3-1 in Chapter 3).

Relations between Photosynthesis and Disease Severity

To examine the effects of disease severity on photosynthesis while controlling for leaf age, measures of leaf A_{sat} were collected on a separate group of leaves of similar age. Measurements were conducted on fully expanded leaves (one to two leaves below the youngest fully expanded leaf) collected from the central two rows in Carver and York at around 25 days after appearance of visual disease symptoms. Symptomatic leaves were selected from non-sprayed plots to determine the reduction of photosynthesis at a given disease severity, whereas asymptomatic leaves were selected from fungicide-sprayed plots on the same day to determine photosynthetic rates under disease-free conditions. Leaflets were selected across a range of LLS severities to quantify the relationship between host genotype photosynthesis and disease severity. Relative leaf A_{sat} was calculated as the A_{sat} of the LLS infected leaflet relative to the average of asymptomatic leaflets, collected on the same day.

Leaf light response curves, CO_2 response curves, and measures of dark-adapted chlorophyll fluorescence were made using the LI-6400XT photosynthesis system, to evaluate determinants of photosynthetic metabolism. Across the two years, another separate group of 20-30 leaflets of similar age, but differing disease severities (including asymptomatic leaflets) were selected for each cultivar as described above and analyzed for these variables. For light response curves, leaf CO_2 assimilation rate (A) was

measured at eight light levels ranging from 0 to 1800 μmol m^{-2} s^{-1}, which were used to estimate quantum efficiency of CO_2 assimilation (ϕ_{CO2}) as described by Boote et al. (1985). Plots of A vs. substomatal CO_2 concentration (c_i) were used to estimate maximum carboxylation velocity of Rubisco ($V_{c,max}$) using the equations of Farquhar et al. (1980). Data were corrected to 25°C using published temperature responses (Long and Bernacchi, 2003). Dark-adapted chlorophyll fluorescence was measured using LI-6400 fitted with a leaf chamber fluorometer (Model LI6400-40), after dark adapting the leaves for around 30 min. Dark-adapted efficiency of Photosystem II (PSII) photochemistry (F_v/F_m) was determined on asymptomatic and symptomatic leaves.

Chlorophyll extraction was performed as described by Inskeep and Bloom (1985). Leaf discs (area = 2 cm^2; diam. = 1.6 cm) were excised from asymptomatic (n = 4) and symptomatic diseased leaflets (n = 25). Leaf discs were placed in glass tubes wrapped in aluminum foil with 5 mL of N, N-dimethylformamide. The tubes were capped to minimize evaporation and then placed on a horizontal shaker for 72 h to extract leaf chlorophyll. Following extraction, 1 ml of solution was placed in a quartz cuvette and absorbance was measured at 647 and 664.5 nm with a spectrophotometer (BW-VIS Model, StellarNet Inc., Tampa, FL). Chlorophyll content (μg cm^{-2}) was calculated for each leaf sample using the equations described by Inskeep and Bloom (1985).

Following the photosynthetic measurements, leaflets were detached and taken to the laboratory. All harvested leaves (and leaf discs) were scanned at 300 dpi using a flatbed scanner (Microtek ScanMaker 5800, Microtek Int. Inc., Industrial Park Hsinchu, Taiwan) and stored as .tiff files. Leaf images were processed using ASSESS ver 2.0 image analysis software (American Phytopathological Society, St. Paul, MN) to give the

percent disease severity (Figure B-2, Erickson et al., 2003). Disease severity was defined as the fraction of total necrotic lesion area to total leaf area.

Data Analysis

On a given sampling date, significant cultivar differences between disease severity and A_{sat} on tagged leaf cohorts through time were analyzed using analysis of variance in the GLIMMIX procedure of SAS (SAS Institute, 2009). Relations between relative Asat and disease severity were analyzed using a non-linear model, $y = (1 - x)^{\beta}$, as described by Bastiaans (1991). The non-linear mixed effects (NLME) library of R (R Development Core Team, 2008) was used to estimate β and assess significant differences ($P < 0.05$) in β values between cultivars. Since, β values did not differ by year within cultivar ($P > 0.05$), the years were pooled for β estimates. Comparisons of the impact of LLS between two peanut cultivars on other photosynthetic variables ($V_{c,max}$, ϕ_{CO2}, F_v/F_m, and chlorophyll) were made under three disease categories: no disease (0% disease severity), low disease (0-15% disease severity), and high disease (15-30% disease severity). Categories were chosen to get approximately equal numbers (n = 10 - 20) in each of three pooled categories (Kumudini et al., 2010).

Results

Disease Severity and A_{sat} on Tagged Leaf Cohorts over Time

Late leaf spot epidemics occurred in both years of the study, but appeared earlier in 2009 compared to 2008 (Figure 4-1). Late leaf spot symptoms were first observed visually in the field around 91 and 82 DAP during 2008 and 2009 on both cultivars, respectively. Across all tagged leaf cohorts, average maximum disease severity was 27 and 19% during 2008, and 25 and 12% during 2009 in Carver and York, respectively. Moreover, the time required from leaf tagging to attain these severities was 51 and 63

days during 2008, and 42 and 49 days during 2009 in Carver and York, respectively. Hence, Carver showed more rapid disease progress than York during both years of the study (Figure 4-1).

Initial net carbon assimilation rate of tagged leaves did not differ between cultivars across all the tagged cohorts, averaging 38.3 and 37.9 μmol m^{-2} s^{-1} during 2008 and 2009, respectively. However, A_{sat} declined in all tagged cohorts with increasing leaf cohort age and disease severity (Figure 4-2). Despite differences in disease severity on individual leaf cohorts (Figure 4-1), A_{sat} did not differ between the two cultivars on any given sampling date (except at 107 DAP in leaves tagged at 65 DAP during 2009) throughout the growing season.

Relations between Photosynthesis and Disease Severity

The mean photosynthetic rate of asymptomatic leaves averaged 36.0 μmol m^{-2} s^{-1} and did not differ ($P > 0.05$) between the two cultivars. In both cultivars, relative A_{sat} was strongly and negatively related to increasing disease severity (Figure 4-3). The β value obtained for both cultivars was greater than 1.0 ($P < 0.001$), indicating that the photosynthetic impairment extended beyond the necrotic lesion area. In addition, the β value for York ($\beta = 4.6 \pm 0.15$) was significantly greater ($P < 0.05$) than the β value for Carver ($\beta = 3.6 \pm 0.12$), which indicated a greater reduction in A_{sat} at a given disease severity for York compared to Carver. Notably, β values were the same for a given cultivar across both years of the study.

The cultivars did not differ in $V_{c,max}$ for asymptomatic leaves with an average value of 148.7 μmol m^{-2} s^{-1}, indicating similar carboxylation capacity between the two cultivars (Figure 4-4A). Maximum carboxylation velocity of Rubisco declined rapidly with increasing disease severity. Moreover, this decline was greater in the more resistant

cultivar York compared to the cultivar with poor resistance to LLS (Carver). In fact, $V_{c,max}$ of leaves with low disease severity (< 15%) was reduced by 62 and 84% compared to asymptomatic leaves in Carver and York, respectively. For leaves with high disease severity, the decline in $V_{c,max}$ was 78 and 83% compared to asymptomatic leaves in Carver and York, respectively. Therefore, these results indicated that the reduction in $V_{c,max}$ was much greater proportionately than disease severity and also that the cultivars differed in $V_{c,max}$ more at relatively low disease severities compared to higher disease severities.

Quantum efficiency of CO_2 assimilation (ϕ_{CO2}) of asymptomatic leaves also did not differ between the cultivars (0.064 mol CO_2 mol^{-1} quanta) and declined rapidly with disease severity (Figure 4-4B). However, the relative decline was not as severe as the decline in $V_{c,max}$. Quantum efficiency of leaves with low disease severity declined 23 and 43% compared to asymptomatic leaves in Carver and York, respectively. For leaves with high disease severity, this decline was 39 and 53% in Carver and York, respectively. Thus, the overall decline in $\phi_{CO2}$was greater in York compared to Carver. As was observed in $V_{c,max}$, differences among cultivars in their response to disease severity was most evident in low disease category.

While $V_{c,max}$ and ϕ_{CO2} were strongly affected at low disease severities, dark-adapted maximum efficiency of PSII (F_v/F_m) was relatively unaffected in both cultivars at low disease (Figure 4-4C). In fact, even at high disease severities, Carver and York showed a similar reduction in F_v/F_m of about 13% compared to their asymptomatic counterparts.

Finally, leaf chlorophyll content also declined with increasing disease severity (Figure 4-4D). Mean chlorophyll content of asymptomatic leaves averaged 53.3 μg cm^{-2} and did not differ between the two cultivars. However, the reduction in chlorophyll at low disease severities was greater in York (43%) than Carver (26%). For leaves with high disease severity, this decline was 32 and 50% in Carver and York, respectively. Thus, the overall decline in chlorophyll content was greater in York compared to Carver.

Discussion

The present study demonstrated differing disease severity and photosynthetic responses to LLS between two peanut cultivars. Despite differences in disease severity, similar reductions in A_{sat} with leaf age were observed for all leaf cohorts across the two cultivars. These similar reductions in carbon assimilation between the cultivars could be explained in part by differing photosynthetic reductions in the leaf tissue beyond the necrotic leaf area (i.e., different β values). A greater decline in A_{sat} at a given disease severity was observed for York compared to Carver. Consistent with a greater β value for York, greater reductions in chlorophyll, ϕ_{CO2}, and especially $V_{c,max}$ were observed in York compared to Carver.

Delayed LLS disease progress, including reduced necrotic lesion area and/or defoliation has been demonstrated in many recently released cultivars (Tillman et al., 2008; Woodward et al., 2008). For example, for the same two cultivars used in the present study, disease severity, based on area under canopy lesion area curve, was reduced by 30% in the York cultivar, which has greater resistance to LLS (Chapter 3). After the onset of LLS, disease severity on tagged leaves differed on all sampling dates between the two cultivars (Figure 4-1), which was attributed to the slower progress of

necrotic lesion area in York compared to Carver. Less necrotic area can lead to less sporulation for this polycyclic disease. Moreover, slow disease progress in resistant genotypes has been related to a longer latent period for the fungus (Chiteka et al., 1988).

Although the cultivars differed in disease severity, leaf photosynthetic behavior was similar with leaf age during the growing season. Light-saturated net assimilation rates did not differ between cultivars at the first sampling date after tagging, which indicated similar photosynthetic potential between the cultivars. Throughout the season, A_{sat} of tagged leaves declined with leaf age (Figure 4-2), attributable to both natural senescence (Guinn and Brummett, 1993) and the incidence of LLS (Bourgeois and Boote, 1992; Nogues et al., 2002). Bassanezi et al. (2002) suggested that reduction in photosynthesis associated with disease was related to the trophic relationship of the pathogen with the host. Values of β reported for some biotrophic pathosystems include 1.3 and 2.2 for *Uromyces appendiculatus* (Pers.:Pers) Unger on common bean (*Phaseolus vulgaris* L.) (Lopes and Berger, 2001 and Bassanezi et al., 2001, respectively) and 2.3 for *Phakospora pachyrhizi* Syd. & P. Syd. on soybean (*Glycine max* L. Merr.) (Kumudini et al., 2010). Thus, β values of biotrophs have generally been equal to or very close to one, indicating minimal effects of biotrophic pathogens on photosynthesis beyond the visual lesion areas of the leaf. However, β values reported for hemibiotrophic pathosystems have generally been greater than 1.0 and often greater than 3.0. For example, Erickson et al. (2003) reported 6.1 for *Marssonina brunnea* f. sp. brunnea on poplar (*Populus* spp.), Bassanezi et al. (2001) reported 3.8 for *Phaeoisariopsis griseola* (Sacc.) Ferr. on common bean, and Roloff et al. (2004) found

values of 2.8 and 3.1 for *Septoria albopunctata* Cooke on *Vaccinium* spp. Consequently, greater reductions in photosynthesis beyond the visual lesion area seem to be more common with hemibiotrophs compared to biotrophs.

Equally important to the magnitude of β, is whether cultivars differ in β, as this could be key for cultivar improvement and for modeling assimilation responses to disease. In this study, intraspecific differences in the photosynthetic response to LLS disease severity were found. Disease induced reduction in A_{sat} was more severe (β = 4.6) in the cultivar with a higher level of resistance to LLS compared to the cultivar with a low level of resistance (β = 3.6). Bourgeois and Boote (1992) reported a decline in relative photosynthesis with disease severity of 4.0 (comparable to β) for peanut (cv. Florunner). Thus, genotypic variability in β is likely to extend beyond the peanut cultivars examined here, and intraspecific variation in β values has been reported in other pathosystems as well (Erickson et al. 2003; Zhang et al., 2009), but is often not seen in biotrophs (Bassanezi et al., 2001; Kumudini et al., 2010). In this study, low disease severity was associated with a high β parameter, but Zhang et al. (2009) reported low β values associated with low disease severities in *Populus cathayana* Rehd., indicating that β is not necessarily inversely correlated with disease severity. The greater reduction in photosynthetic capacity beyond the necrotic lesion area in York compared to Carver at a given disease severity could help to explain the similar reductions in canopy carbon assimilation through time between the two cultivars and their respective yield responses in the field (Chapter 3), despite differences in disease severities as explained earlier.

Reductions in Asat with increasing disease severity were also associated with reductions in $V_{c,max}$, ϕ_{CO2}, F_v/F_m, and chlorophyll. Results indicated that reductions in all the determinants of photosynthetic metabolism were much greater proportionally than the necrotic lesion area except for F_v/F_m. Among the various determinants of photosynthesis studied, $V_{c,max}$ showed the greatest relative decline, especially in the low disease category. Nogues et al. (2002) also concluded that decreased $V_{c,max}$ was likely the primary determinant underlying the decline in A_{sat} of tomato (*Lycopersicon esculentum* Mill.) infected by *Fusarium oxysporum* f.sp. lycopersici. Moreover, the cultivars exhibited a differential response in $V_{c,max}$ to disease severity, which could potentially explain the differing photosynthetic response between the cultivars. Taken together, these results indicated that the resistant cultivar, York, was unable to sustain photosynthesis in response to LLS despite its ability to slow the progression of disease (i.e. resistance).

In conclusion, this study demonstrates the variability in photosynthetic response to LLS among cultivars with differing resistance levels. York, the cultivar with the higher level of resistance to LLS showed more photosynthetic impairment beyond the necrotic lesion area at a given disease severity compared to Carver, the cultivar with a poorer level of resistance. Possible mechanisms responsible for this greater photosynthetic impairment in York included a reduction in carboxylation velocity of Rubisco. These findings were attributed in part to a lack of photosynthetic tolerance to LLS in the more resistant cultivar York. These results have potential implications in our efforts for selecting improved cultivars and predicting growth and yield responses of new peanut cultivars to leaf spot. For example, combining visual disease ratings with physiological

measures of the β parameter could result in the identification and selection of cultivars with slow disease progress (e.g., like York) and relatively low β parameters (e.g., like Carver), which could contribute to reduced yield loss due to LLS, especially under low fungicide input production. Since collection of β values is relatively intensive, spectral methods that detect chlorophyll as a surrogate for β represents a future research need along with evaluation of a greater diversity of cultivars.

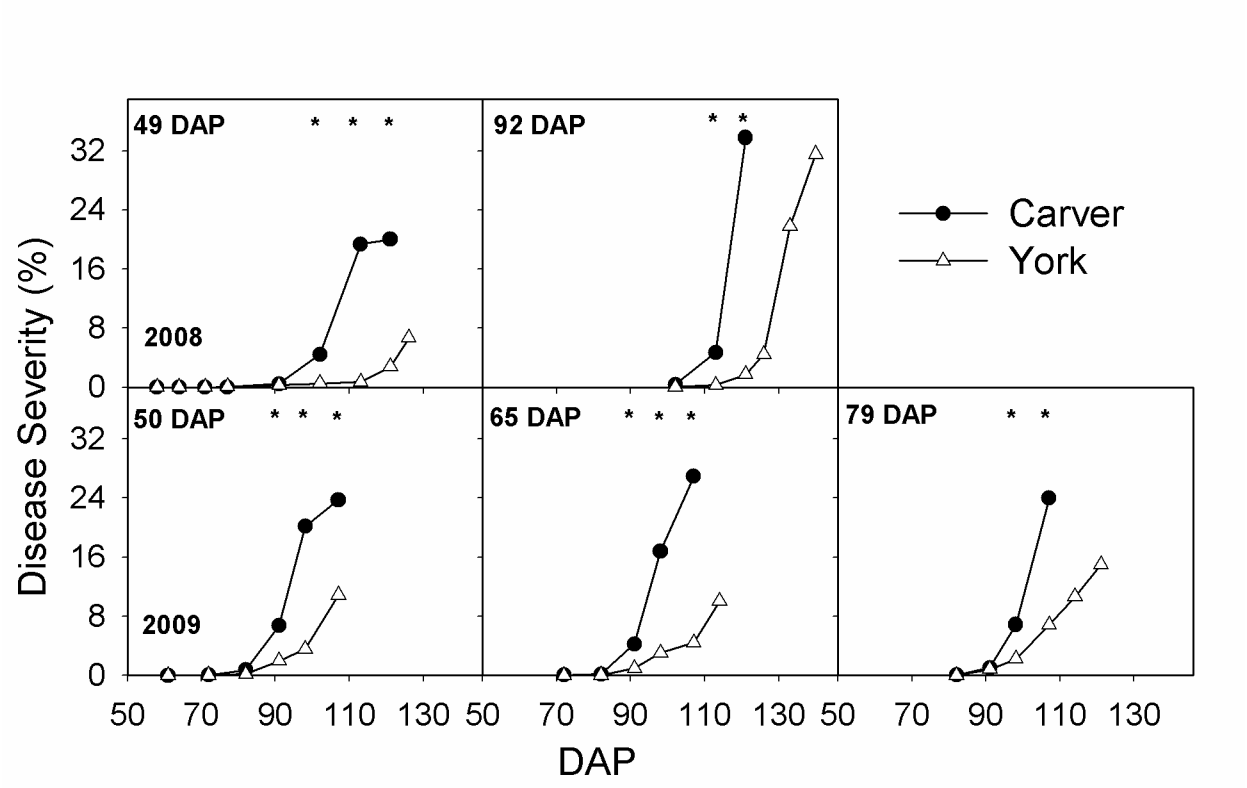

Figure 4-1. Progress of late leaf spot (LLS) severity (percent necrotic lesion area) on the individual leaf cohorts during 2008 and 2009 growing seasons for the two peanut cultivars Carver and York, grown under no-fungicide application conditions. Leaf cohorts were tagged at 49 and 92 days after planting (DAP) during 2008 and at 50, 65, and 79 DAP during 2009 growing seasons, respectively. Significant differences ($P < 0.05$) between cultivars for each time point are indicated by an asterisk.

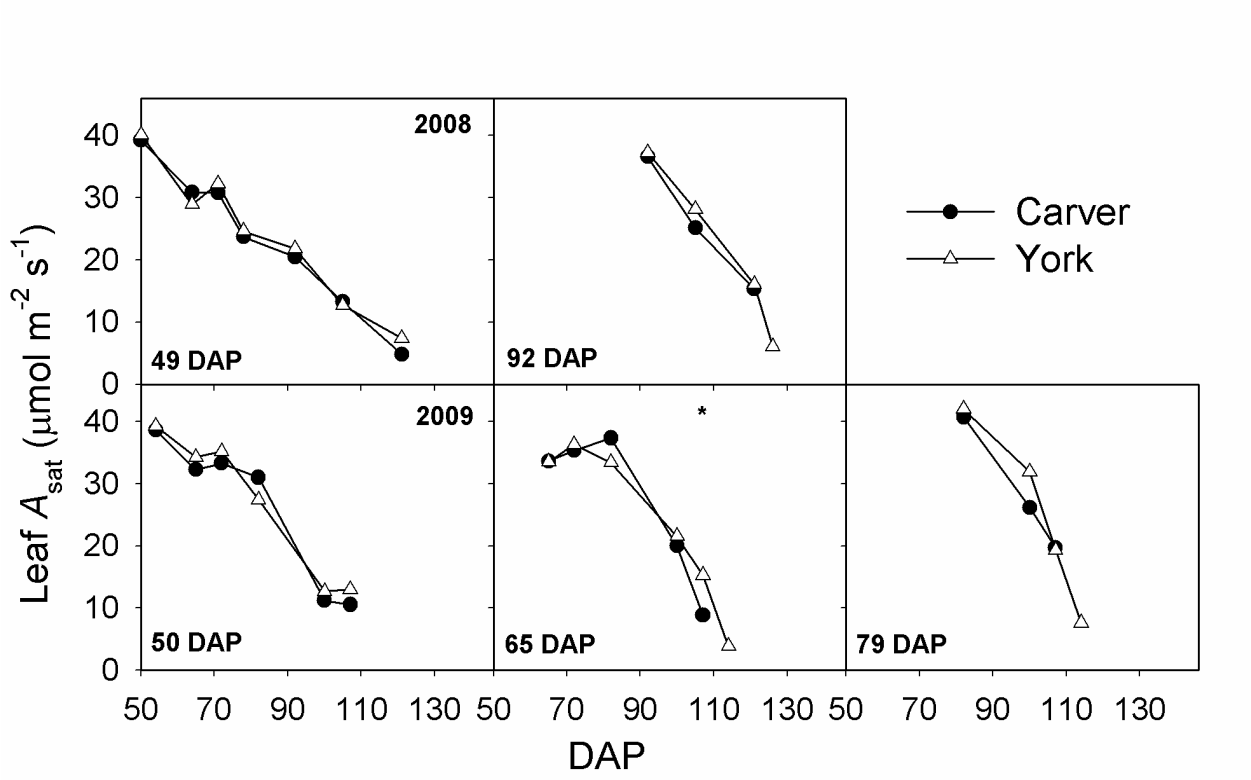

Figure 4-2. Light-saturated CO_2 assimilation rate (Leaf A_{sat}) of individual leaf cohorts during 2008 and 2009 growing seasons for the two peanut cultivars Carver and York, grown under no-fungicide application conditions. Leaf cohorts were tagged at 49 and 92 days after planting (DAP) during 2008 and at 50, 65, and 79 DAP during 2009 growing seasons, respectively. Significant differences ($P < 0.05$) between cultivars for each time point are indicated by an asterisk.

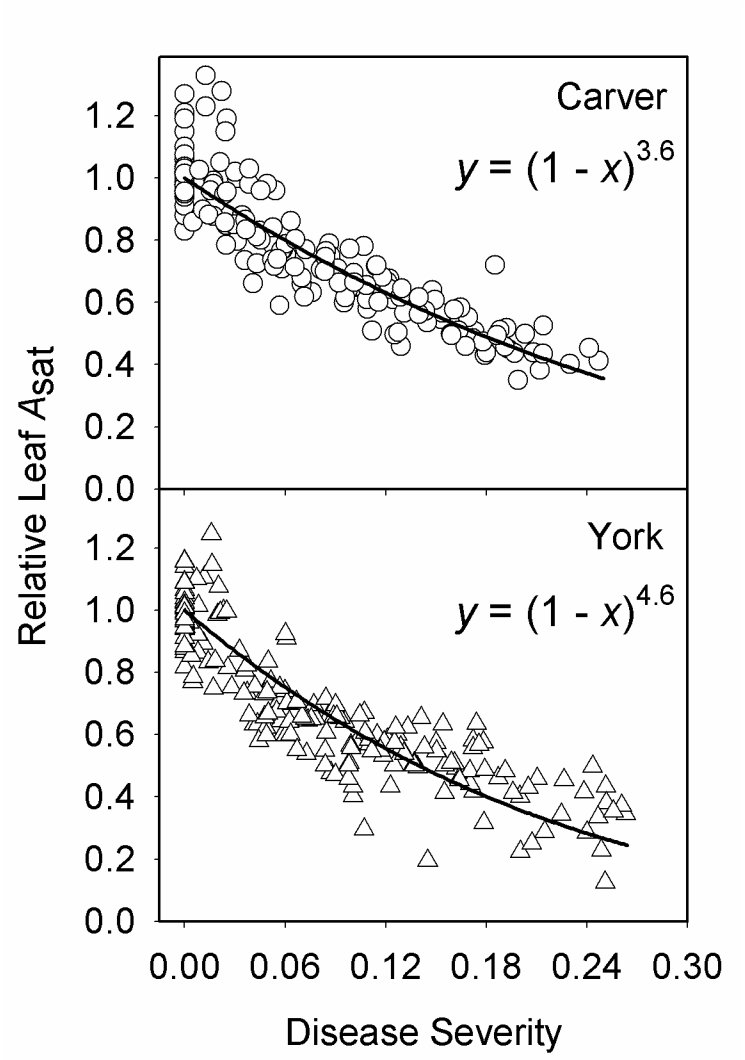

Figure 4-3. Relative light-saturated leaf CO_2 assimilation rate (Relative leaf A_{sat}) in relation to disease severity (fraction necrotic lesion area) for two peanut cultivars Carver and York. β parameter for York (4.6, n = 183) was greater (P < 0.05) compared to that of Carver (3.6, n = 160).

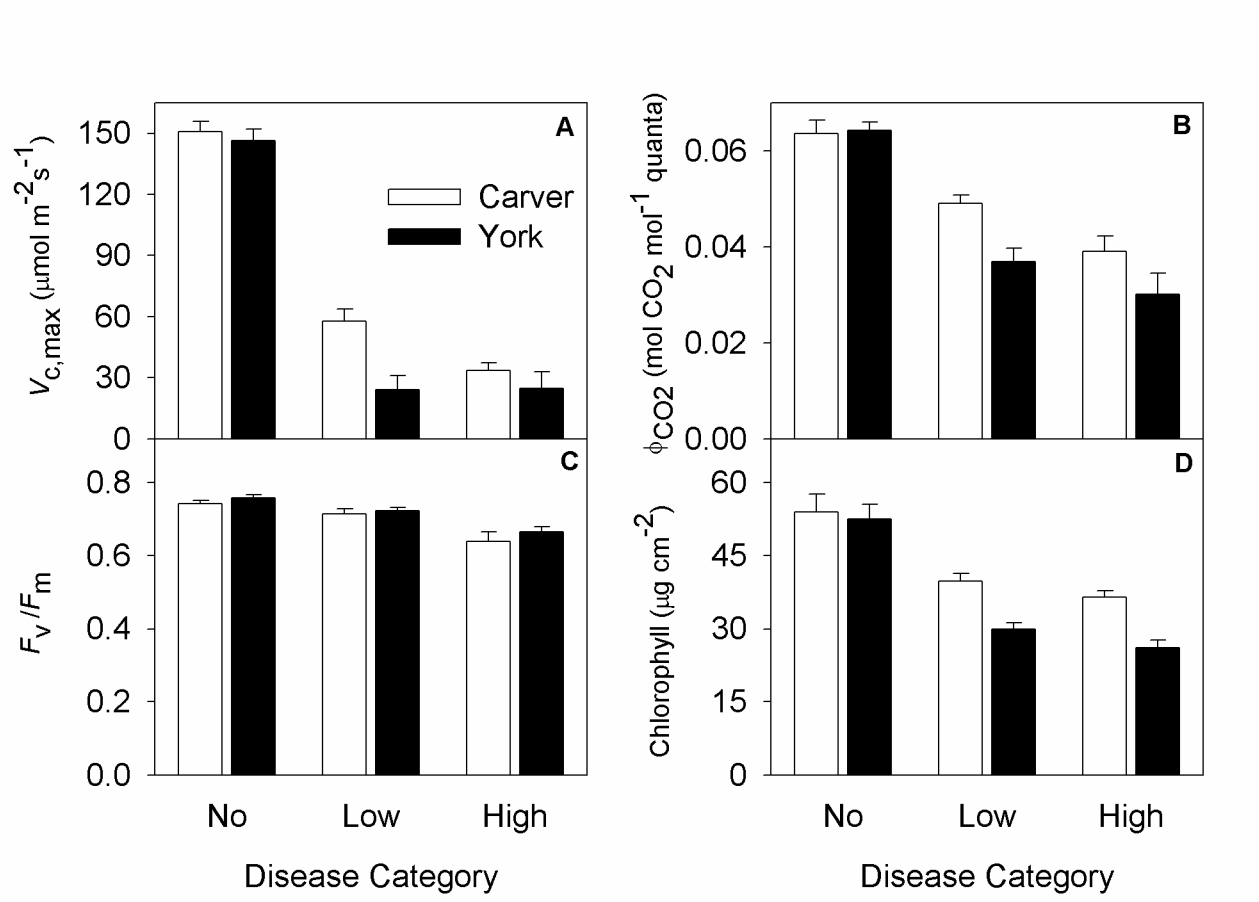

Figure 4-4. Changes in (*A*) maximum carboxylation velocity of Rubisco ($V_{c,max}$), (B) quantum efficiency of CO_2 assimilation (ϕ_{CO2}), (C) dark-adapted maximum efficiency of PSII photochemistry (F_v/F_m), and (D) chlorophyll content of leaves at no (0% disease severity), low (0 - 15% disease severity), and high (15 - 30% disease severity) disease categories for two peanut cultivars, Carver and York.

CHAPTER 5
USING THE CROPGRO-PEANUT MODEL TO SIMULATE GROWTH AND YIELD IN PEANUT CULTIVARS WITH VARIABLE RESISTANCE LEVELS TO LATE LEAF SPOT

Abstract

Late leaf spot (LLS) caused by *Cercosporidium personatum* (Berk. & Curt.) Deighton leads to significant reductions in peanut (*Arachis hypogaea* L.) yield worldwide. This study was conducted to determine whether LLS effects on defoliation and photosynthesis can be incorporated into the CROPGRO-Peanut model to simulate growth and yield reductions in peanut cultivars. Field experiments were conducted in 2008 and 2009 to collect data on the effects of LLS on biomass accumulation and partitioning, leaf necrosis and defoliation, and total canopy photosynthesis (TCP) in peanut cultivars with more (York) and less (Carver) quantitative resistance to LLS. After incorporating LLS damage as percent defoliation and necrotic area (with cultivar specific *β* value), the model accurately simulated crop growth and development for both cultivars despite different disease dynamics. Simulated leaf, total crop, and pod yield values were in good agreement with measured data. Agreement between measured and simulated TCP values indicated correct crop C balance. A modification in the model to directly reduce leaf photosynthesis and quantum efficiency resulted in improved simulations of LLS effects on growth and yield of both cultivars. Correlations among measured defoliation and necrotic area with disease ratings indicated that visual disease ratings could be successfully used to estimate necrosis and defoliation and to correctly simulate LLS-induced reductions in growth and yield. Results indicated that the CROPGRO-Peanut model has adequate capabilities to simulate LLS effects on growth and yield in peanut cultivars with differing levels of resistance to LLS when inputs on

canopy necrotic area and defoliation are provided, which could be used to improve model predictions to help reduce fungicide use and improve cultivar development.

Background

In the southeastern USA, early leaf spot (caused by *Cercospora arachidicola* S. Hori), and late leaf spot [caused by *Cercosporidium personatum* (Berk. & Curt.) Deighton] are among the most widespread and damaging foliar diseases of peanut (*Arachis hypogaea* L.) (Nutter Jr. and Shokes, 1995). Yield reductions associated with these foliar diseases are related to premature loss of green leaf area (by necrotic tissue and defoliation) and reduction of leaf photosynthetic capacity (Pixley et al., 1990a; Bourgeois and Boote, 1992; Chapter 4). Late leaf spot (LLS) can cause yield losses of up to 50% in some cases (Pixley et al., 1990a; Bourgeois et al., 1991). Current crop protection strategies rely largely on fungicide applications, cultural practices, and resistant cultivars. But economic and environmental concerns of fungicides have increased the demand for improved management strategies based on minimizing crop losses rather than minimizing disease outbreaks. Decision support systems that can predict yield losses rather than controlling the outbreak of disease and hence provide improved disease management options are required to meet this demand.

Crop models are important tools to evaluate growth and yield losses due to various biotic and abiotic stresses (Boote et al., 1983a; Naab et al., 2004; Timsina et al., 2007). Models that have been used to predict the impact of foliar diseases on yield have generally incorporated the disease effects on defoliation and photosynthesis (Batchelor et al., 1993; Boote et al 1993; Teng et al., 1998). Disease-induced defoliation has been incorporated into the models by simply reducing the leaf area (Batchelor et al., 1993; Williams and Boote, 1995); however the reduction in leaf photosynthesis due to disease

is more complex to model. The impact of LLS on leaf photosynthesis has been shown to be greater than can be accounted for by the visual lesion area (Bourgeois and Boote, 1992). In order to relate reductions in leaf photosynthesis to visual lesion area, Bastiaans (1991) proposed a relatively simple model, $y = (1- x)^{\beta}$, where y is the relative photosynthetic rate of a diseased leaf compared to that of an asymptomatic leaf, x is the measured visual lesion area, and β describes the relationship between virtual and visual lesion area. The virtual area represents loss of photosynthetic capacity beyond the visual lesion area. Thus, β indicates whether the effect of disease on photosynthesis is higher ($\beta > 1$), lower ($\beta < 1$), or equal ($\beta = 1$) to that accounted for by the measured visual lesion area. Several studies have incorporated this parameter into crop growth models to estimate growth and yield losses due to biotrophic pathogens at the canopy level (Bastiaans 1993; Bassanezi et al., 2001; Robert et al., 2004; Bancal et al., 2007) but all these researchers used a single β value for all cultivars. However, the photosynthetic response (β parameter) can differ among various genotypes in their response to a pathogen (Erickson et al., 2003; Zhang et al., 2009; Chapter 4). In these cases, using a cultivar specific β parameter in the crop model should achieve better predictions of disease-induced reductions in carbon assimilation, growth, and yield. Quantifying the effects of LLS on peanut cultivars with variable levels of resistance and inclusion of the effect on photosynthesis in yield loss models are of great importance for a more complete understanding of growth and yield responses to diseases and should increase the accuracy of yield loss estimates.

The CROPGRO-Peanut model (Boote et al., 1998a, 1998b) is a process-oriented mechanistic crop growth model which considers crop carbon balance, crop and soil N

balance, and soil water balance at the process level. This model has coupling points and procedures for entering pest damage to simulate growth and yield reductions associated with foliar pathogens like LLS (Batchelor et al., 1993; Boote et al., 1993). The primary impacts of disease are simulated as defoliation; however the impacts of virtual lesion area are currently simulated by simply defoliating more leaf area (hence zero photosynthesis on that area) rather than creating direct impact at the leaf level photosynthesis. This subroutine has been tested by some previous studies (Naab et al., 2004; Adomou et al., 2005) to simulate LLS effects on peanut growth and yield. However, these studies did not include measured data on leaf necrosis and/or defoliation required by the disease subroutine in the model. Either visual ICRISAT ratings were linearly regressed against necrosis values of 0 to 9% (based on data from Bourgeois et al., 1991) to obtain hypothesized necrosis values (Adomou et al., 2005) or variable defoliation and necrosis values were used to mimic leaf weight loss (Naab et al., 2004). Moreover, these studies were conducted only on cultivars lacking any known level of resistance to LLS and not on cultivars differing in their resistance levels to LLS. This signifies the need of having reliable disease assessment methods to provide accurate assessment of disease effects in cultivars with differing disease resistance levels, which could then be used as input to crop growth models and as evaluation methods in breeding programs.

The overall objective of this study was therefore to evaluate the CROPGRO-Peanut model for its ability to simulate the impacts of LLS on growth and yield reductions in peanut cultivars with variable resistance levels when inputs on necrosis and defoliation were provided. The different physiological response (β values) among

peanut cultivars to LLS was incorporated into the model to test its ability to accurately simulate reductions in growth and yield. In addition, the model was modified to directly impact the light-saturated leaf photosynthetic rate (A_{sat}) and quantum efficiency of CO_2 assimilation (QE) depending on the percent necrotic area, as observed in the physiological data on disease effects from the field experiments (Chapter 4). Multiple disease assessment methods were also analyzed to determine the best method to estimate necrosis and defoliation, or to derive them from visual ratings (e.g. Florida 1-10 scale). Thus, these objectives represented a step towards improved model simulation of LLS-induced growth and yield losses that when coupled with a disease simulation model or subroutine will contribute to reduced fungicide use and improved peanut cultivar development.

Materials and Methods

Experimental Site and Design

Data were obtained from field experiments conducted during the 2008 and 2009 growing seasons at the Plant Science Research and Education Unit in Citra, Florida (29°23'60'' N, 82°12'0'' W) to simulate growth and yield reductions associated with LLS epidemics on peanut cultivars of differing resistance using CROPGRO-Peanut model. These experiments were part of a larger study conducted to quantify the growth and yield losses and underlying physiological determinants due to LLS in peanut cultivars with variable levels of resistance. The soil at the experimental site was Gainesville loamy sand (hyperthermic, coated Typic Quartzipsamments). The experiment was a two by two factorial arranged in a randomized complete block design with four replications. Cultivar and fungicide application were treated as fixed effects. Two cultivars were selected for differences in resistance to LLS: Carver (Gorbet, 2006) has poor resistance

to LLS; while York (Gorbet and Tillman, 2011) has moderate resistance to LLS (Tillman et al., 2008). Fungicide application included: (i) no fungicide application and (ii) an industry-standard fungicide schedule (Table 3-1) applied on a 14-d interval commencing from approximately 40 days after planting (DAP).

Sowing occurred during the latter part of the recommended planting window for North Central Florida on May 20 in 2008 and May 27 in 2009 to maximize LLS pressure (Wright et al., 2006). Each plot consisted of 6 rows spaced 0.91 m apart and 4.6 m long. Seeds were sown at a rate of 17-20 seeds per meter row using a conventional planter. Standard management practices for irrigated peanut were employed during both years (Wright et al., 2006) to manage the crop as described in Chapter 3.

Measures of Growth and Yield

Starting 35 DAP, a 61-cm section of row was harvested randomly from the outer two rows in each plot at approximately biweekly intervals to measure growth and partitioning. A representative subsample plant was selected from each harvested sample (Bourgeois et al., 1991; Pixley el al., 1990b). The remaining harvested sample was immediately oven dried for 72 h at 60^{o}C and subsequently weighed. Leaflets and pods were separated from all subsamples and then leaves, stems, and pods were oven dried to a constant weight. Stem, leaf, and pod dry weights (DW) were determined for the entire sample by multiplying their respective fractions of the subsample times the total weight of the harvested sample.

A 61-cm section of row was selected randomly from the outer two rows starting around 35 DAP to measure total canopy photosynthesis (TCP), using a 91 by 61 cm aluminum-frame mylar chamber and a portable photosynthesis system (LICOR LI-6200, Li-Cor Inc., Lincoln, NE). Total canopy photosynthesis was calculated by adding the

absolute dark respiration (measured under dark conditions) to the measured carbon exchange rate under full sunlight conditions, as described in Chapter 3.

Measures of Disease Injury

Occurrence and severity of late leaf spot were assessed visually based on the widely used Florida 1-10 scale (Table B-1, Chiteka et al., 1988; Woodward et al., 2008; Woodward et al., 2010). Values of 1 to 4 indicate increasing leaf spot incidence on leaflets within the lower or upper canopy, but with no defoliation. Ratings from 4 to 10 are associated with increasing levels of defoliation (Chiteka et al., 1988). Ratings began when visual symptoms first appeared and continued every 7-10 days until harvest. Microscopic examination of lesions on leaflets indicated that *C. personatum* was the dominant pathogen in both years. Spotted wilt (caused by Tomato spotted wilt virus) and white mold (caused by *Sclerotium rolfsii* Sacc.) were not observed in the field plots during either growing season.

Canopy defoliation and necrosis, the components that make up the Florida scale ratings, were also measured objectively by other methods throughout the growing season to compare to the more subjective Florida 1 to 10 scale assessment. To determine canopy lesion area, forty leaflets were randomly selected from the subsample plant. All leaflets were scanned at 300 dpi using a flatbed scanner (Microtek ScanMaker 5800, Microtek Int. Inc., Industrial Park Hsinchu, Taiwan) and stored as .tiff files. Leaf images were processed using ASSESS ver 2.0 image analysis software (American Phytopathological Society, St. Paul, MN) to give the percent necrotic area (Figure B-2, Erickson et al., 2003). Leaf hue was used by the program to distinguish necrotic lesion area.

To calculate percent canopy defoliation, different methods were employed as follows: (i) Loss of leaf DW from peak leaf weight was used to calculate percent defoliation for each treatment. After the occurrence of peak leaf DW, percent defoliation was calculated for that treatment on a given day as the ratio of the difference between peak leaf DW and leaf DW on the given sampling day to the peak DW observed (Pixley et al., 1990b). (ii) Number of total nodes and nodes with missing leaflets were counted on the main-stem of each subsample plant (Pixley et al., 1990b; Adomou et al., 2005). Percent defoliation was calculated as the ratio of missing to total nodes for each plant. The first six and eight nodes were subsequently not considered in counting for Carver and York, respectively, to account for the differences in non-disease induced leaf senescence which varied with life cycle and branch formation from lower nodes between the two cultivars (Adomou et al., 2005). The intercept of the linear relation between missing nodes on the main-stem and defoliation (based on leaf weight loss from peak weight) was used as a starting point to reach the number of nodes to exclude (Figure A-1), and was further optimized by the best CROPGRO-Peanut model fit against the actual leaf DW data. (iii) Number of total nodes and nodes with missing leaflets were also counted on the four dominant lateral branches and percent defoliation was determined as described above. The percent defoliation values obtained through these methods (Tables A-1 and A-2) were used as input for model simulations in file T under the header PCLA as described later.

To obtain estimates of necrosis and defoliation as a function of visual disease ratings, Florida 1-10 scale ratings were correlated to necrosis and to defoliation by comparing the slopes and intercepts of the relations using generalized least squares

procedure of the nlme library of R (R development core team, 2008). Data were combined if slope and intercept of the given relation was not significant ($P > 0.05$).

Description of the CROPGRO-Peanut Model

The CROPGRO-Peanut model is a mechanistic, process oriented model designed to simulate growth and development on a daily basis using crop C, crop and soil N, and soil water balances (Boote et al, 1998a, 1998b). The code is modular and generic, such that crop-specific parameters were removed from the code and placed into read-in species, ecotype, and cultivars files. Crop development includes the rates of vegetative and reproductive development (expressed as physiological days as a function of temperature, photoperiod, water, and N deficit) that governs dry matter partitioning to plant organs over time. Crop N balance includes daily soil N uptake, N_2 fixation, N mobilization from vegetative tissues, and N loss from abscised parts. Soil water balance includes infiltration of irrigation and rainfall, runoff, drainage, root uptake, soil evaporation, and plant transpiration. Crop C balance includes daily photosynthesis, growth and maintenance respiration, conversion and condensation of C to crop tissues, and C losses to abscised parts.

The CROPGRO-Peanut model computes canopy photosynthesis at hourly time steps using leaf-level photosynthesis and hedge-row light interception (Boote and Pickering, 1994). This approach is more mechanistic and responsive to row spacing and plant density. Absorption of direct and diffused irradiance by sunlit versus shaded leaves is computed based upon canopy height and width, row direction, leaf angle, latitude of the site, day of year and time of day along with the predicted LAI (Boote and Pickering, 1994). Photosynthesis of sunlit and shaded leaves is computed using the asymptotic exponential light response equation:

$$A = A_{\max} \times [1.0 - \exp(-QE \times PPFD / A_{\max})] \quad (5\text{-}1)$$

Where A is leaf CO_2 assimilation rate (μmol m^{-2} s^{-1}), A_{max} is the light-saturated A (defined at 30°C, 350 μmol CO_2 mol^{-1} air), QE is the quantum efficiency of the leaf (referenced at the same conditions as A_{max}), and PPFD is the photosynthetic photon flux density. Both Amax and QE can be adjusted based on the following factors:

$$A_{\max} = A_{\max,g} \times f(Temp) \times f(Chill) \times f(CO_2) \times f(leafN) \times f(SLW) \times f(\beta_{\max}) \quad (5\text{-}2)$$

$$QE = QE_g \times f(CO_2) \times f(leafN) \times f(\beta_{QE}) \quad (5\text{-}3)$$

Where $A_{max,g}$ is a cultivar-specific coefficient, QE_g is a C-3 species-specific coefficient; and *f* represents 0-1 adjustment function based on temperature of given day (temp), minimum temperature of previous night (Chill), atmospheric CO_2 concentration (CO_2), leaf N concentration (leafN), leaf thickness (specific leaf weight, SLW), and disease impact (β_{max} or β_{QE}). Both β_{max} and β_{QE} are not present in the default model and have been introduced into the model as a part of this study (as explained later) and are calculated as:

$$\beta_{\max} or \beta_{QE} = (1 - PDLA)^{\beta} \quad (5\text{-}4)$$

Where PDLA is the fraction necrotic lesion area and β represents the relationship between virtual and visual lesion area.

Hourly canopy photosynthesis on a land area basis is computed by multiplying the photosynthetic rates for the sunlit and shaded leaves by their respective LAIs. The hourly rates are integrated over the 24 hr period to yield the total daily gross photosynthesis. Since CROPGRO is a source-driven crop model, correctly predicting canopy assimilation is important. Finally, growth of new tissues depends on daily

carbohydrate availability, partitioning to various tissues, and respiration costs of tissue synthesis.

CROPGRO-Peanut Model Inputs

Daily weather data (i.e. maximum and minimum temperature, rainfall, and solar radiation) were obtained on site from the Florida Automated Weather Network (FAWN, 2011) for both growing seasons. Data on site (latitude, longitude, and elevation), crop management practices (e.g. sowing date, spacing, plant population), and soil type and properties were provided as input to the model as well. All data were entered in the standard file formats (*.PNX, *.PNA, *.PNT, *.WTH, and SOIL.SOL) needed for execution of the CROPGRO-Peanut model in DSSAT ver 4.5 (Jones et al., 2003; Hoogenboom et al., 2009). Data on disease-induced necrosis and defoliation were also entered in the model input file to simulate disease damage as explained later.

Procedure for Calibration of Genetic Coefficients

The CROPGRO-Peanut model requires genetic coefficients that describe crop phenology, vegetative growth traits, and reproductive growth traits unique to a given cultivar (Boote et al., 1998b). Carver, the cultivar with poor resistance to LLS, is phenotypically similar to Florunner; thus the genetic coefficients of Florunner available in DSSAT version 4.5 were chosen as the starting point for calibration. For York, genetic coefficients of Southern Runner were chosen initially, as this is a longer cycle cultivar that also has moderate levels of resistance to LLS. These coefficients were modified slightly by comparing the simulated phenology, time series growth and yield with observed data from the fungicide-sprayed plots only up to mid-season, following the procedures outlined by Boote (1999). The extent of changes in coefficients was small

(Table 5-1) and this exercise was done for more accurate prediction of timing of vegetative and reproductive growth. Model calibration was not the point of the study.

Procedure for Simulating Disease Effects

The effects of LLS on growth and yield of peanut were simulated by entering the measured levels of percent necrosis due to disease and associated percent leaf defoliation for the corresponding day of year into the crop performance file (File T) of DSSAT, under the headers of PDLA (necrosis percentage) and PCLA (defoliation percentage, Tables A-1 and A-2). The default model code is designed to read this file and interpolates between dates to create leaf area loss (also leaf mass and N loss) (Batchelor et al., 1993; Boote et al., 1993). The effect of necrosis (PDLA) in the model is amplified by the presence of virtual lesion area (Bastiaans 1991; Erickson et al., 2003; Chapter 4). The model uses a virtual lesion effect of 4.0 irrespective of the cultivar used (estimated for Florunner by Bourgeois and Boote, 1992), meaning there is an effective four-unit decrease in photosynthesizing leaf area for every unit of necrotic disease area. The model accounts for this effect by defoliating more leaf area based on this virtual effect. The model then runs the remainder of the season with reduced leaf area (and mass), resulting in reduced light interception, canopy photosynthesis, and yield.

In Chapter 4, the virtual lesion effect (β value) differed for the two cultivars used in this study. To account for this, β values of 3.6 and 4.6 were used for Carver and York, respectively in all the simulations conducted in this study. Moreover, a modification was made in the model, whereby the effect of necrosis was placed directly on A_{sat} (eq. 5-2) and QE (eq. 5-3) of single leaf, to correspond directly to the physiological data collected in the field study. So, instead of defoliating more leaf area as a result of necrosis and virtual lesion (as in the default model by assuming linear relationship between necrotic

area and effective leaf area), this modification to the model caused a cultivar-specific reduction in A_{sat} and QE, assuming that the necrotic spot was directly in the photosynthetic area (Figure 5-1, eq. 5-1 to 5-4). Simulation runs conducted by this routine are referred to as modified model simulations.

Statistical Evaluation of Model Performance

Evaluation of model performance was conducted using root mean square error (RMSE) and the Willmott (1981, 1982) index of agreement (D-index). These statistical indicators were computed from observed and simulated variables (e.g. leaf mass, pod mass, and total crop biomass). The RMSE reflects the magnitude of the root mean sum of square differences between the predicted (*P*) and observed (*O*) values over time and is calculated as:

$$RMSE = \sqrt{\frac{\sum_{i=1}^{n}(P_i - O_i)^2}{n}}$$

The D-index is a descriptive index that measures dispersion of the simulated and observed data, calculated as:

$$D - index = 1 - \left[\frac{\sum_{i=1}^{n}(P_i - O_i)^2}{\sum_{i=1}^{n}\left(| P_i - \overline{O} | + | O_i - \overline{O} | \right)^2} \right]$$

Where *n* is the total number of observations, P_i is the predicted value for the *i*th measurement, O_i is the observed value for the *i*th measurement, and $\overline{O}$ is the overall mean of the observed values. A model performs well when the RMSE approaches zero and the D-index is close to 1.0.

Results and Discussion

Leaf Weight and Leaf Area Simulations

The temporal changes in measured leaf dry weight along with the simulated values are presented in Figures 5-2 and 5-3. The model (with no defoliation function) predicted leaf dry weight with good accuracy until around 95 and 85 DAP during 2008 and 2009 respectively for Carver and York, after which the model consistently over-predicted the leaf dry matter accumulation. This occurred as leaf dry weight began to decline due to LLS disease around that time and the model was unable to simulate those impacts (the model assumes no disease-induced necrosis and defoliation). This decline was observed even in fungicide-sprayed plots (Figures 5-2 and 5-3), indicating that the 14-d calendar based fungicide program did not achieve 100% disease control. To account for the LLS induced damage, the defoliation function in the model was used as explained earlier, whereby defoliation was input over time as calculated by different methods. Inclusion of the defoliation function resulted in improved simulations of leaf weight (Figures 5-2 and 5-3, Table 5-2). Using leaf weight loss as defoliation function resulted in good agreement between simulated and measured values, as shown by high D-index values (close to 1) and low RMSE values (Table 5-2). However, destructive growth sampling is required to collect leaf weight loss data, which is very labor intensive and time consuming. Counting main stem nodes and/or branch nodes to calculate defoliation is comparatively easier, but resulted in overestimation of leaf weight loss due to LLS (Figures 5-2 and 5-3). This could occur due to natural leaf senescence occurring on lower nodes, and can be accounted by subtracting the first few nodes on the main stem (Adomou et al., 2005). The first six and eight nodes on main stem in Carver and York were excluded to calculate defoliation, resulting in good agreement between

simulated and measured values (Figures 5-2 and 5-3, Table 5-2). More nodes were excluded in York to account for higher leaf senescence of lower nodes which could occur due to longer life cycle and more branching from lower nodes in York. Adomou et al. (2005) did not count the first four nodes in their study conducted on short duration cultivars. These results showed the importance of a proper "missing node" method to estimate defoliation caused by LLS in peanut. The D-index values for leaf weight obtained in this study (0.96-1.00) after accounting for defoliation (using leaf weight loss) were higher than those obtained in other studies using the same method (0.68-0.84, Adomou et al., 2005).

Overall, simulated LAI followed the same trend as described above for leaf mass (data not shown). In both cultivars, simulated LAI was close to measured values until around 90 and 80 DAP during 2008 and 2009 respectively without using the defoliation function, and further values were improved by including the disease-induced defoliation. Because LAI in the model is a function of both leaf mass and specific leaf area (SLA), this indicated that the SLA simulations were also good.

Simulations of Total Biomass and Pod Weight

The temporal changes in observed total biomass and pod weight along with the predicted values are presented in Figures 5-4 and 5-5. Without the disease function, the model simulated total biomass and pod weight accurately up to around 110 DAP for both cultivars after which disease reductions on leaf area and assimilation become important. Thereafter in the model over predicted both total biomass and pod yield resulting in high RMSE and only moderate d-index (Tables 5-3 and 5-4). To account for LLS-induced damage, both measured necrosis and defoliation (calculated from leaf weight loss as explained in the previous section) were used as a disease function to run

the simulations. This resulted in good agreement between the simulated and measured total biomass and pod weight across the growing season, as shown by D-index values approaching 1.0 and low RMSE values (Tables 5-3 and 5-4). This was evident under both fungicide-sprayed and non-sprayed conditions during both years of the study. Reductions in total biomass and pod yield even under fungicide-sprayed conditions corroborates other studies which showed less than optimal control of LLS with standard program (Monfort et al., 2004; Woodward et al., 2010). Naab et al. (2004) also showed good agreement between simulated and measured total biomass (D-index = 0.98) and pod yield (D-index = 0.97) after entering hypothesized necrosis and defoliation values to mimic the leaf weight loss.

All model simulations for York were run using a different virtual lesion effect in York (β = 4.6) and Carver (β = 3.6). Table 5-5 shows improvement in RMSE values for total biomass and pod yield simulations in York with the use of higher β value for York. Although the improvement in RMSE value is small, this shows some importance of using cultivar-specific virtual lesion effect in modeling growth and yield losses where intraspecific differences in the physiological response to disease are seen. Moreover, use of cultivar specific β value (3.6 in Carver and 4.6 in York) resulted in improved predictions in 2009 when disease incidence occurred earlier in the season and progressed faster compared to 2008 (data not shown). This shows that the importance of the use of different β values may depend on disease severity and the factors that contribute to greater disease severity such as time of onset, weather conditions, etc. In other cases where intraspecific variability does not exist or is minimal, it is possible to

use same β value for cultivars (Robert et al., 2006; Bancal et al., 2007; Kumudini et al., 2010).

Simulations showed that the slope of biomass accumulation rose smoothly after a short lag phase early in the vegetative growth period. This lag phase was longer in York compared to Carver. Both measured and simulated values indicated a peak dry weight of around 10,000 kg ha^{-1} in both cultivars. Thereafter, the simulated dry matter accumulation showed a decline in both cultivars which occurred due to LLS incidence. Comparison of the seasonal patterns of simulated pod weight among the cultivars also showed later initiation of pod fill, slower pod growth rate, and longer duration of pod fill in York compared to Carver.

Simulations of Canopy Photosynthesis

Figure 5-6 shows model simulations of TCP, with and without the disease function. Without the necrosis and defoliation inputs, the model simulations were poor with low D-index and high RMSE values. Canopy photosynthesis simulations improved with the use of the disease function, except for the sharp drops caused by cloudy days (Figure 5-6, Table 5-6). On cloudy days, TCP was actually measured under full sun (late morning and between cloud breaks), whereas simulated photosynthesis rates were based on total daily solar radiations and hence were low on cloudy days. Agreement between simulated and measured TCP values indicated correct crop C balance in the model. Decline in TCP in fungicide-sprayed plots after the incidence of LLS again indicated less than 100% control of disease by calendar-based fungicide schedule. Reductions in TCP due to LLS have also been observed by Bourgeois and Boote (1992) while working with Florunner cultivar in peanut.

Simulations with the Modified Model

The modified model was used to directly impact A_{sat} and QE (Chapter 4) to account for the virtual lesion area (β value). Figure 5-7 shows model simulations of total biomass and pod weight under non-sprayed conditions for 2009, with the default and modified model. The modified model resulted in some improvement in the simulations as shown by the increase in D-index and decrease in RMSE values compared to the default model simulations for both years (Table 5-7 and 5-8). Simulations for all treatments in 2008 and under fungicide-sprayed conditions for 2009 are not shown as the trends were similar to the ones shown in Figure 5-7. Improvement in model predictions of total crop and pod weight showed the importance of more mechanistically including the effects of LLS on A_{sat} and QE in modeling LLS impacts.

Estimating Disease Induced Percent Necrosis and Defoliation from Florida 1-10 Scale

Percent canopy necrosis was positively related to Florida 1-10 visual rating scale ($P < 0.001$). The slope of the relation between percent necrosis (measured with scan system) and Florida 1-10 scale was not affected ($P > 0.05$) by cultivar, fungicide schedule, or year (Figure 5-8). Adomou et al. 2005 also developed a relationship between percent necrosis and the ICRISAT 1-9 scale which is similar to Florida scale. However, their relation was developed by linearly regressing necrosis from zero at ICRISAT score of 1 to a maximum necrosis of 9% at ICRISAT score of 8, based on necrosis data from Bourgeois et al. (1991). The relation established in this study is more reliable and rigorously tested as it was developed from actual scan-measured data on two peanut cultivars under fungicide-sprayed and non-sprayed conditions across two years of study. This relationship between the visual rating scale and necrosis did not

differ among treatments in this study and can be used to derive percent necrosis from visual rating scale where measured data on necrosis is not available.

Percent canopy defoliation (calculated from leaf weight loss) was also positively related to Florida 1-10 scale ($P < 0.001$). The slope of the relation between defoliation and Florida 1-10 scale was not affected by fungicide schedule or year, however, it was influenced by the cultivar used (Figure 5-9). This showed that the Florida scale ratings were not consistent for defoliation among the two cultivars. These results indicated that a cultivar-specific relation is required to derive defoliation from visual rating scale.

Simulations Using Estimated Necrosis and Defoliation from Florida 1-10 Scale

Simulations conducted using percent necrosis and defoliation inputs derived from visual disease ratings (Florida 1-10 scale) were in good agreement with the simulated values using measured disease data (Figure 5-10; Tables 5-9 and 5-10). The temporal dynamics of total crop biomass and pod weight were consistent between both simulations and were in good agreement with the measured data as well, as shown by D-index values approaching 1.0 and RMSE values approaching zero. These results indicated that input for disease function can be derived from the less labor intensive and less time consuming visual disease scouting methods rather than relying on intensive measurements for disease-induced necrosis and defoliation. The D-index values for pod weight (0.95-0.99) obtained in this study were higher than the ones (0.69-0.87) obtained by Adomou et al. (2005) while using actual defoliation and estimated necrosis data. This indicated more accuracy of the relations developed in this study in estimating quantitative disease damage from visual ratings.

Conclusions

The CROPGRO-Peanut model can be used to simulate the influence of foliar diseases (e.g. late leaf spot) on photosynthesis, growth, partitioning, and yield reductions in peanut cultivars with differing levels of resistance to LLS when inputs on percent canopy necrotic area and defoliation are provided. Estimating defoliation from main stem nodes worked well, but obtaining the appropriate starting point (node 7 in Carver and node 9 in York) for disease-induced defoliation is important. Significant relations between Florida 1-10 visual rating scale and measured necrosis and defoliation can be used to simulate LLS-induced growth and yield reductions where detailed sampling on disease damage is not conducted. This approach of entering LLS damage based on scouting information can be extended to other important foliar diseases of peanut and other legumes to simulate disease-induced growth and yield reductions.

The adjustments made to the model code were relatively minor but resulted in improved predictions (10% over prediction of final pod yield by the modified model compared to 14% by the default model across all treatments) of the effect of LLS on growth and development of peanut. It is recommended that the next version of the CROPGRO model should include the changes to simulate effects of necrotic area directly on leaf photosynthetic traits. Use of cultivar-specific β parameter was warranted as it resulted in improved simulations of growth and yield. The sensitivity of the model to percent necrosis (PDLA) and β parameter among different cultivars needs further investigation. Future model development should include an independent disease simulator which can predict LLS-induced necrosis and defoliation based on weather conditions. This would allow improved predictions of the impacts of foliar diseases like

LLS on growth and yield and hence result in reduced fungicide use and improved cultivar development without relying on the disease damage inputs from scouting.

Table 5-1. Genetic coefficients of the cultivars Carver, Florunner, York, and Southern Runner used for model simulations.

		Cultivar			
Genetic coefficient	Abbreviation	Carver	Florunner	York	S. Run‡
Time from emergence to flower appearance, pd†	EM - FL	20.2	21.2	24.0	22.9
Time from beginning flower to beginning pod, pd	FL - SH	9.2	9.2	10.6	9.2
Time from beginning flower to beginning seed, pd	FL - SD	18.8	18.8	20.4	18.2
Time from beginning seed to maturity, pd	SD - PM	74.3	74.3	76.0	82.6
Time from beginning flower to end of leaf expansion, pd	FL - LF	85	88	86	91
Maximum leaf photosynthetic rate, mg CO_2 m^{-2} s^{-1}	LFMAX	1.40	1.40	1.33	1.30
Specific leaf area, cm^2 g^{-1}	SLAVR	232	260	232	265
Maximum size of full leaf, cm^2	SIZELF	18	18	14	17
Maximum fraction of daily growth partitioned to seed and shell	XFRT	0.92	0.92	0.83	0.85
Maximum weight per seed, g	WTPSD	0.68	0.69	0.68	0.63
Seed filling duration, pd	SFDUR	42.9	40.0	43.0	40.0
Seeds per pod, no. pod^{-1}	SDPDV	1.72	1.65	1.71	1.65
Time to reach full pod load, pd	PODUR	26	24	32	30

† pd, Photothermal days

‡ S. Run., Southern Runner

Table 5-2. Root mean square error (RMSE) and index of agreement (D-index) values for leaf dry weight for two peanut cultivars grown under fungicide-sprayed (Fung) and non-sprayed (NF) conditions during 2008 and 2009. Defoliation inputs include no defoliation input (No def), defoliation based on leaf weight loss (LW), main stem defoliation (MS), branch defoliation (Branch), and main stem defoliation excluding first six and eight nodes in Carver and York (MS + offset).

			Defoliation input									
Year	Cultivar	Fungicide	No def	LW	MS	Branch	MS+offset	No def	LW	MS	Branch	MS+offset
			----------D-index----------					----------RMSE (kg ha^{-1})----------				
2008	Carver	NF	0.66	1.00	0.99	0.97	1.00	951	112	133	293	108
		Fung	0.67	0.96	0.99	0.95	0.98	822	298	98	277	205
	York	NF	0.60	0.98	0.91	0.86	0.99	1066	235	399	487	184
		Fung	0.68	0.99	0.88	0.78	0.98	892	174	446	621	201
2008	Carver	NF	0.70	0.99	0.93	0.95	0.98	913	196	409	392	238
		Fung	0.66	0.97	0.97	0.93	0.96	834	260	237	356	300
	York	NF	0.61	0.99	0.87	0.84	0.99	1105	184	485	591	170
		Fung	0.71	0.96	0.79	0.65	0.96	807	269	552	809	282

Table 5-3. Root mean square error (RMSE) and index of agreement (D-index) values for total biomass for two peanut cultivars grown under fungicide-sprayed (Fung) and non-sprayed (NF) conditions at Citra, FL during 2008 and 2009. Simulations were conducted without (No dis) and with disease function including necrosis and defoliation (Dis function).

Year	Cultivar	Fungicide	No dis	Dis function	No dis	Dis function
			-------D-index------		--RMSE (Kg ha^{-1})--	
2008	Carver	NF	0.93	0.99	1698	517
		Fung	0.93	0.98	1702	941
	York	NF	0.81	0.95	2968	1357
		Fung	0.90	0.97	2311	1072
2009	Carver	NF	0.92	0.97	1869	1071
		Fung	0.95	0.99	1463	664
	York	NF	0.88	0.98	2517	921
		Fung	0.94	0.98	1838	1091

Table 5-4. Root mean square error (RMSE) and index of agreement (D-index) values for pod weight for two peanut cultivars grown under fungicide-sprayed (Fung) and non-sprayed (NF) conditions at Citra, FL during 2008 and 2009. Simulations were conducted without (No dis) and with disease function including necrosis and defoliation (Dis function).

Year	Cultivar	Fungicide	No dis	Dis function	No dis	Dis function
			-------D-index------		--RMSE (Kg ha^{-1})--	
2008	Carver	NF	0.97	0.99	638	359
		Fung	0.96	0.96	751	700
	York	NF	0.93	0.95	802	622
		Fung	0.97	0.97	522	497
2009	Carver	NF	0.95	0.98	790	522
		Fung	0.98	0.99	596	477
	York	NF	0.95	0.97	824	537
		Fung	0.98	0.99	526	455

Table 5-5. Virtual lesion effect (β value) on root mean square error (RMSE) values for total biomass and pod yield for York grown under fungicide-sprayed (Fung) and non-sprayed (NF) conditions during 2008 and 2009.

Year	Cultivar	Fungicide	β = 3.6	β = 4.6	β = 3.6	β = 4.6
			--Total biomass (Kg ha^{-1})--		--Pod weight (Kg ha^{-1})--	
2008	York	NF	1393	1357	647	622
		Fung	1108	1072	508	497
2009	York	NF	945	921	549	537
		Fung	1106	1091	472	455

Table 5-6. Root mean square error (RMSE) and index of agreement (D-index) values for total canopy photosynthesis (TCP) for two peanut cultivars grown under fungicide-sprayed (Fung) and non-sprayed (NF) conditions during 2008 and 2009. Simulations were conducted without (No dis) and with disease function including necrosis and defoliation (Dis function).

Year	Cultivar	Fungicide	No dis	Dis function	No dis	Dis function
			-------D-index------		--RMSE (mg m^{-2} s^{-1})--	
2008	Carver	NF	0.60	0.89	0.74	0.43
		Fung	0.57	0.78	0.67	0.51
	York	NF	0.71	0.96	0.68	0.27
		Fung	0.71	0.93	0.61	0.31
2009	Carver	NF	0.50	0.89	0.82	0.42
		Fung	0.50	0.71	0.61	0.47
	York	NF	0.64	0.93	0.65	0.32
		Fung	0.70	0.86	0.52	0.36

Table 5-7. Comparison of default vs. modified model for root mean square error (RMSE) and index of agreement (D-index) values for total biomass for two peanut cultivars grown under fungicide-sprayed (Fung) and non-sprayed (NF) conditions during 2008 and 2009.

Year	Cultivar	Fungicide	Default	Modified	Default	Modified
			------D-index------		--RMSE (Kg ha^{-1})--	
2008	Carver	NF	0.99	0.99	517	522
		Fung	0.98	0.98	941	845
	York	NF	0.95	0.96	1357	1194
		Fung	0.97	0.98	1072	930
2009	Carver	NF	0.97	0.97	1071	1122
		Fung	0.99	0.99	664	623
	York	NF	0.98	0.98	921	798
		Fung	0.98	0.98	1091	1007

Table 5-8. Comparison of default vs. modified model for root mean square error (RMSE) and index of agreement (D-index) values for pod weight for two peanut cultivars grown under fungicide-sprayed (Fung) and non-sprayed (NF) conditions during 2008 and 2009.

Year	Cultivar	Fungicide	Default	Modified	Default	Modified
			------D-index------		--RMSE (Kg ha^{-1})--	
2008	Carver	NF	0.99	0.99	359	281
		Fung	0.96	0.97	700	617
	York	NF	0.95	0.96	622	540
		Fung	0.97	0.98	497	423
2009	Carver	NF	0.98	0.97	522	553
		Fung	0.99	0.99	477	435
	York	NF	0.97	0.98	537	482
		Fung	0.99	0.99	455	406

Table 5-9. Statistics of total biomass as simulated by CROPGRO-Peanut model using disease function derived from measured data (Dis fun-msd) and estimated data from Florida 1-10 scale (Dis fun-est) for two peanut cultivars grown under fungicide-sprayed (Fung) and non-sprayed (NF) conditions during 2008 and 2009.

Year	Cultivar	Fungicide	Dis fun-msd	Dis fun-est	Dis fun-msd	Dis fun-est
			------D-index†------		---RMSE‡ (Kg ha^{-1})---	
2008	Carver	NF	0.99	0.99	522	580
		Fung	0.98	0.97	845	1004
	York	NF	0.96	0.94	1194	1427
		Fung	0.98	0.96	930	1242
2009	Carver	NF	0.97	0.96	1122	1171
		Fung	0.99	0.99	623	644
	York	NF	0.98	0.98	798	788
		Fung	0.98	0.98	1007	842

† D-index, index of agreement; ‡ RMSE, root mean square error.

Table 5-10. Statistics of pod yield as simulated by CROPGRO-Peanut model using disease function derived from measured data (Dis fun-msd) and estimated data from Florida 1-10 scale (Dis fun-est) for two peanut cultivars grown under fungicide-sprayed (Fung) and non-sprayed (NF) conditions during 2008 and 2009.

Year	Cultivar	Fungicide	Dis fun-msd	Dis fun-est	Dis fun-msd	Dis fun-est
			-----D-index†-----		---RMSE‡ (Kg ha^{-1})---	
2008	Carver	NF	0.99	0.99	281	297
		Fung	0.97	0.97	617	664
	York	NF	0.96	0.95	540	645
		Fung	0.98	0.97	423	526
2009	Carver	NF	0.97	0.97	553	559
		Fung	0.99	0.99	435	398
	York	NF	0.98	0.98	482	415
		Fung	0.99	0.99	406	330

† D-index, index of agreement; ‡ RMSE, root mean square error.

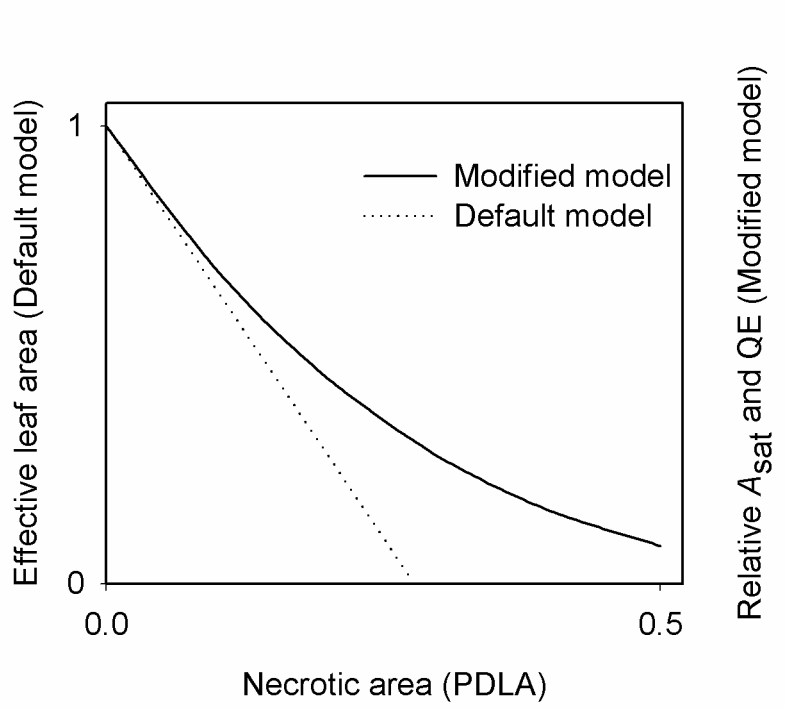

Figure 5-1. Relationship between necrotic area and effective leaf area in the default model and between necrotic area and relative A_{sat} (ratio of photosynthetic rate of diseased leaflet to the average of asymptomatic leaflets) and QE (quantum efficiency of CO_2 assimilation) in the modified model.

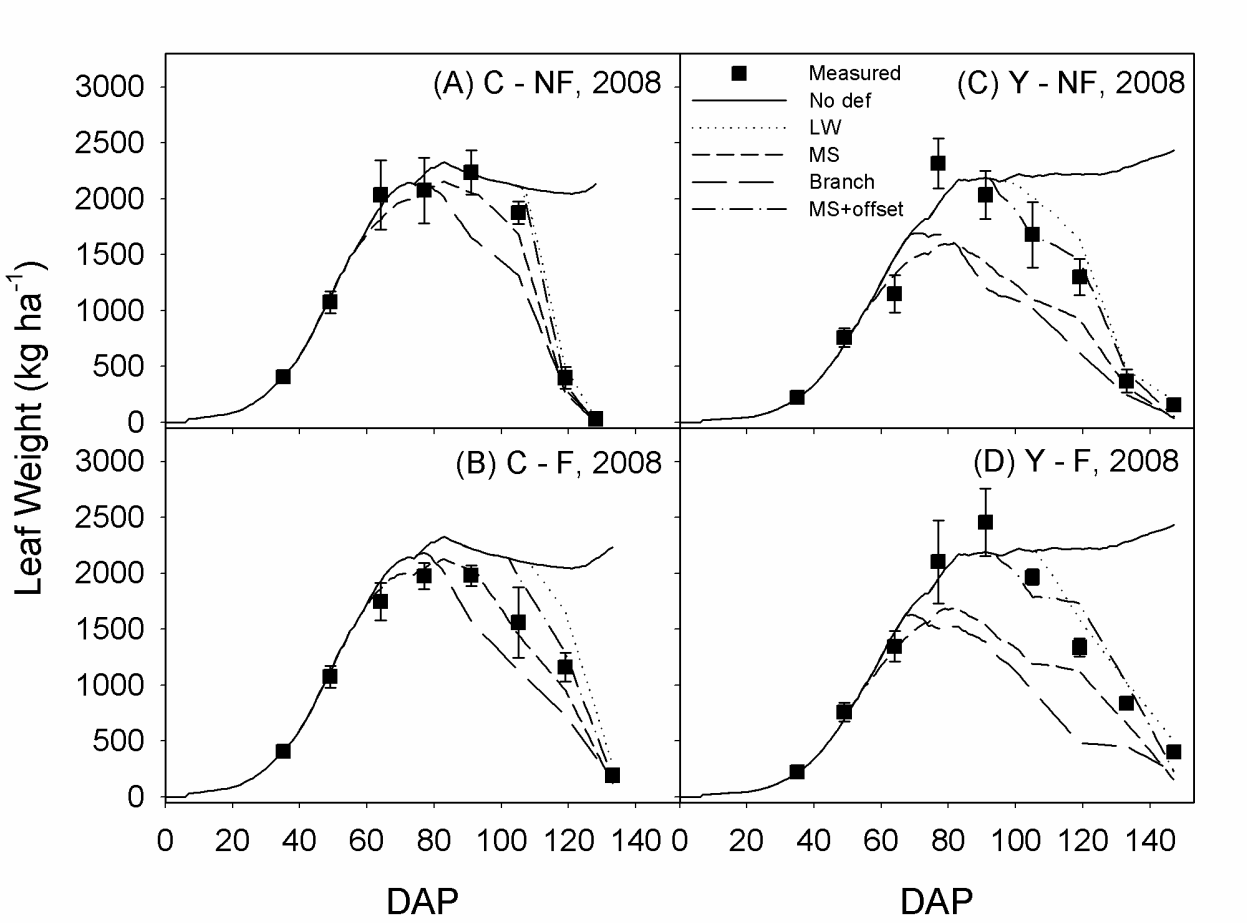

Figure 5-2. Simulated and measured leaf dry weight vs. days after planting (DAP) for peanut cultivars Carver (C) and York (Y) grown under fungicide-sprayed (F) and non-sprayed (NF) conditions during 2008. Symbols represent treatment means (*n*=4). Lines represents simulations based on defoliation inputs including no defoliation input (No def), defoliation based on leaf weight loss (LW), main stem defoliation (MS), branch defoliation (Branch), and main stem defoliation excluding first six and eight nodes in Carver and York (MS + offset). Vertical bars represent ± standard error of the mean.

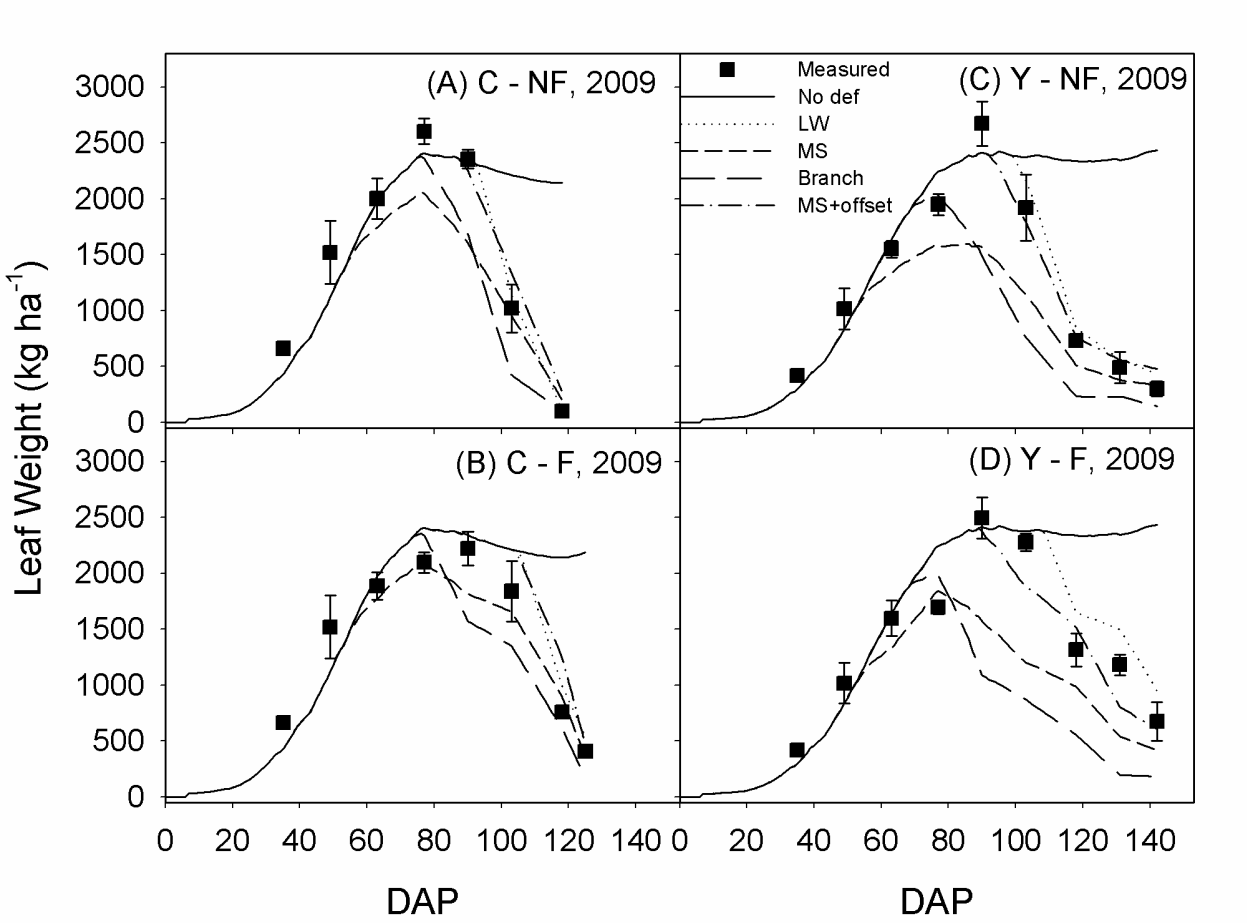

Figure 5-3. Simulated and measured leaf dry weight vs. days after planting (DAP) for peanut cultivars Carver (C) and York (Y) grown under fungicide-sprayed (F) and non-sprayed (NF) conditions during 2009. Symbols represents measured treatment means (*n*=4). Lines represents simulations based on defoliation inputs including no defoliation input (No def), defoliation based on leaf weight loss (LW), main stem defoliation (MS), branch defoliation (Branch), and main stem defoliation excluding first six and eight nodes in Carver and York (MS + offset). Vertical bars represent ± standard error of the mean.

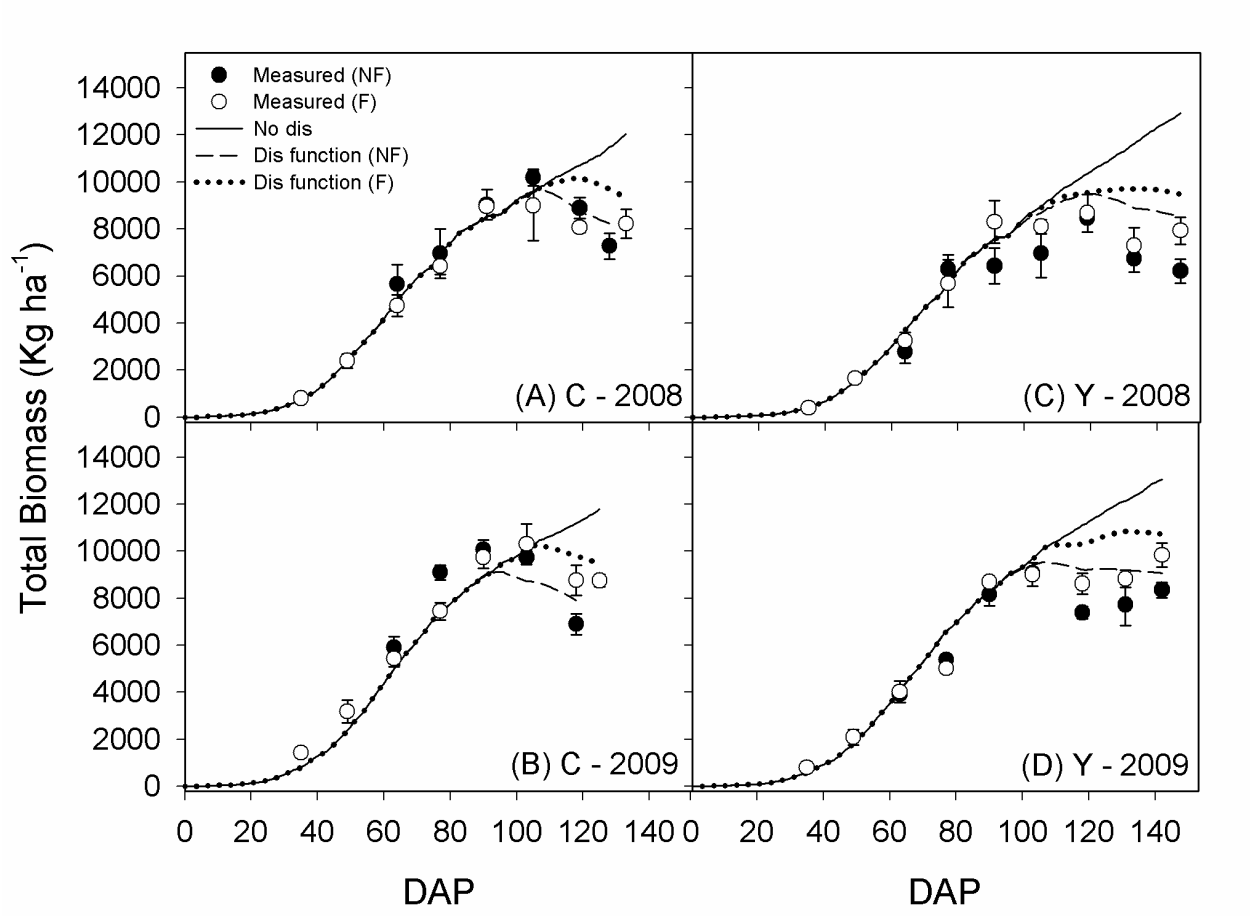

Figure 5-4. Simulated and measured total biomass for peanut cultivars Carver (C) and York (Y) grown under fungicide-sprayed (F) and non-sprayed (NF) conditions at Citra, FL during 2008 and 2009. Symbols represents measured treatment means (*n*=4). Lines represent simulations including no disease function (No dis) and disease function with necrosis and defoliation (Dis function). Vertical bars represent ± standard error of the mean.

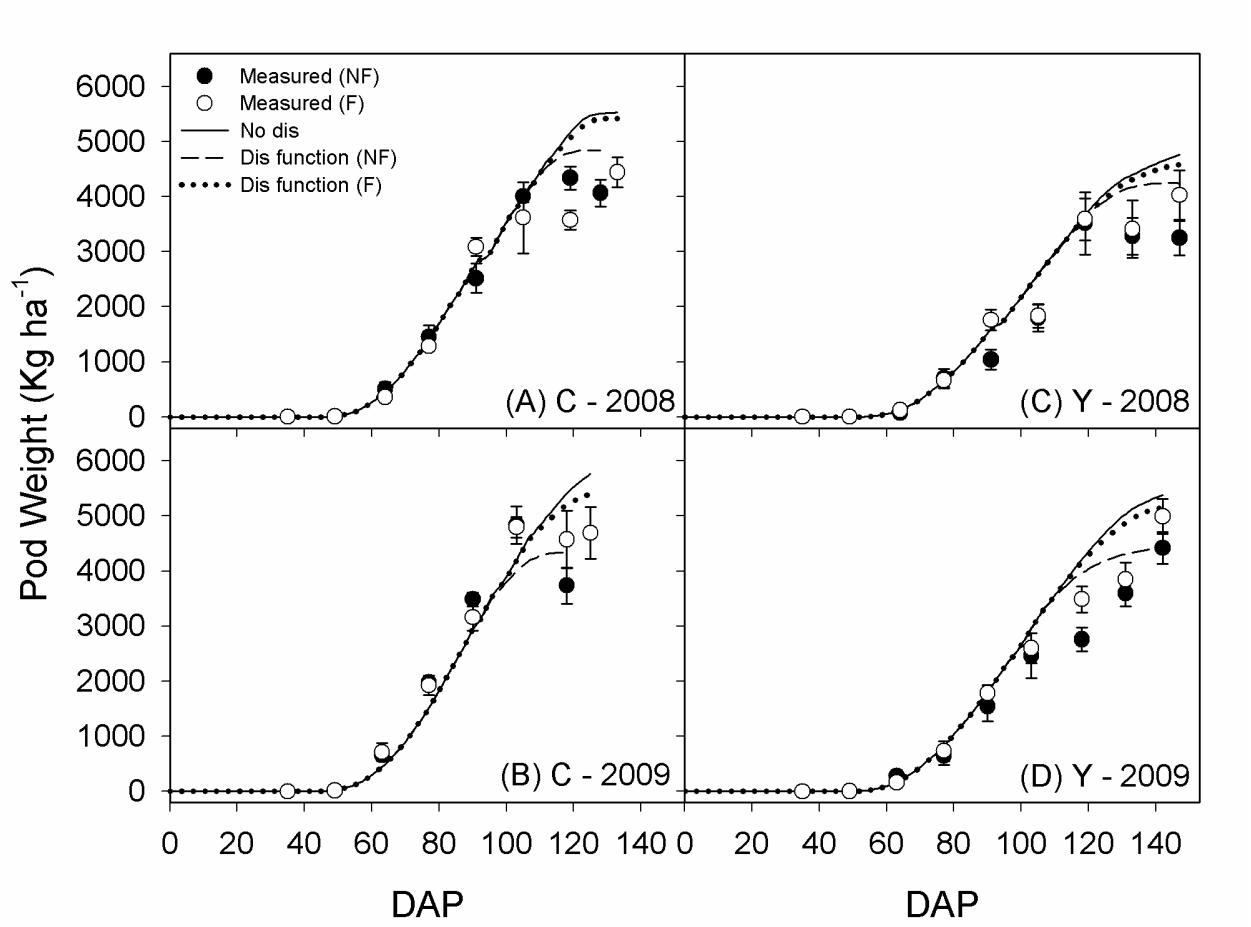

Figure 5-5. Simulated and measured pod weight for peanut cultivars Carver (C) and York (Y) grown under fungicide-sprayed (F) and non-sprayed (NF) conditions at Citra, FL during 2008 and 2009. Symbols represents measured treatment means (*n*=4). Lines represent simulations including no disease function (No dis) and disease function with necrosis and defoliation (Dis function). Vertical bars represent ± standard error of the mean.

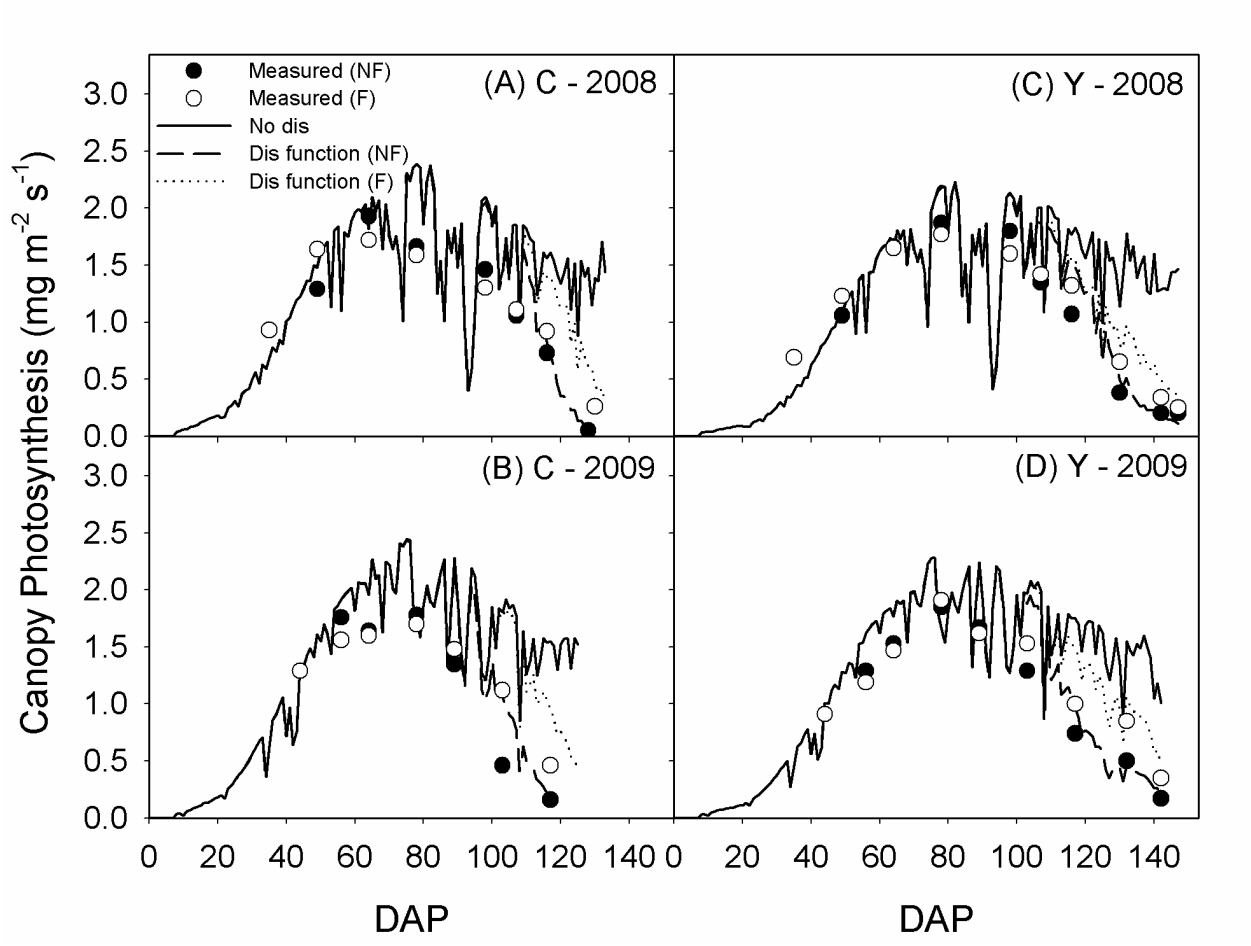

Figure 5-6. Simulated and measured mid-day total canopy photosynthesis (TCP) for peanut cultivars Carver (C) and York (Y) grown under fungicide-sprayed (F) and non-sprayed (NF) conditions at Citra, FL during 2008 and 2009. Symbols represents measured treatment means (*n*=4). Lines represent simulations including no disease function (No dis) and disease function with necrosis and defoliation (Dis function).

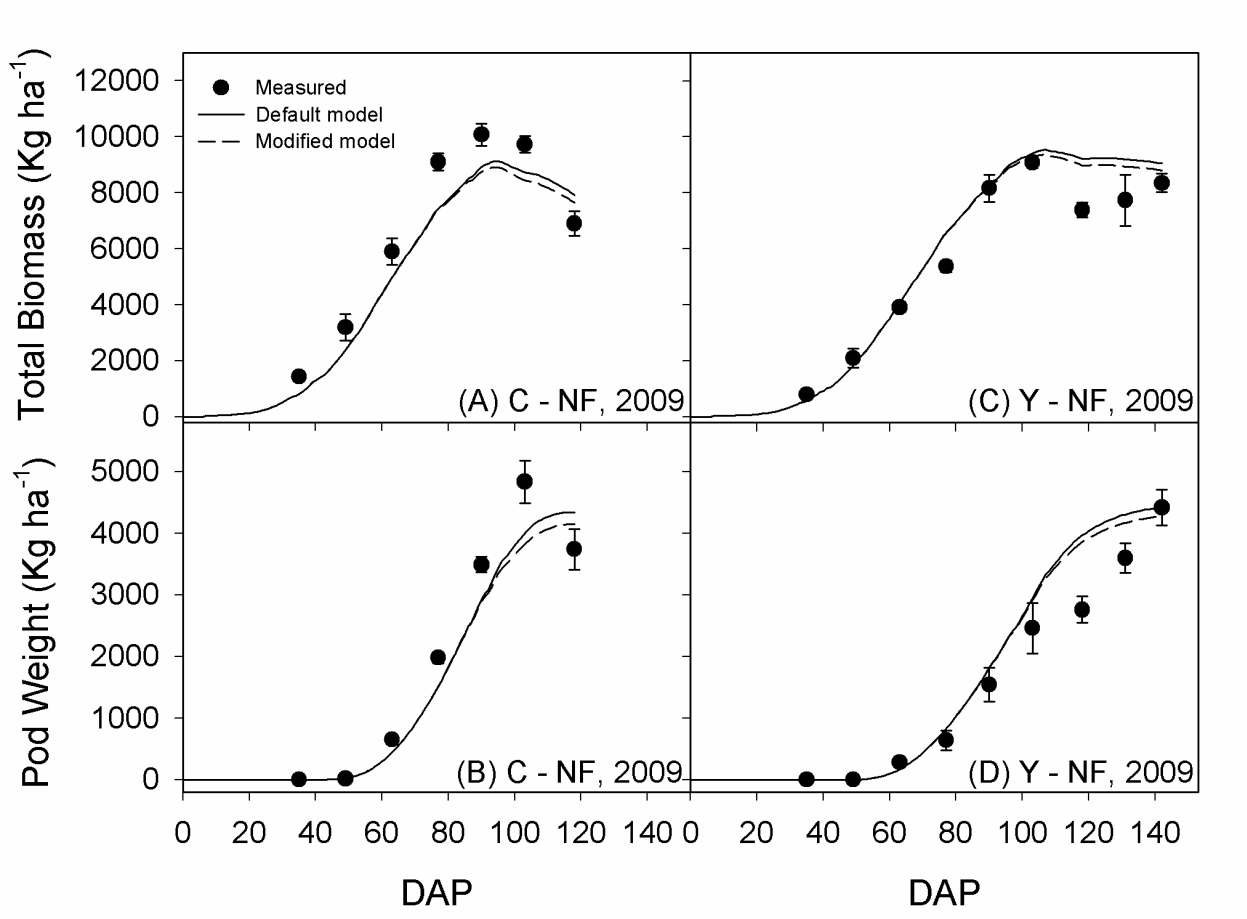

Figure 5-7. Simulated and measured total biomass and pod weight for peanut cultivars Carver (C) and York (Y) grown under no-fungicide application (NF) conditions during 2009. Symbols represents measured treatment means (*n*=4). Lines represents simulations with default model routine (Default model) and modified model routine (Modified model). Vertical bars represent ± standard error of the mean.

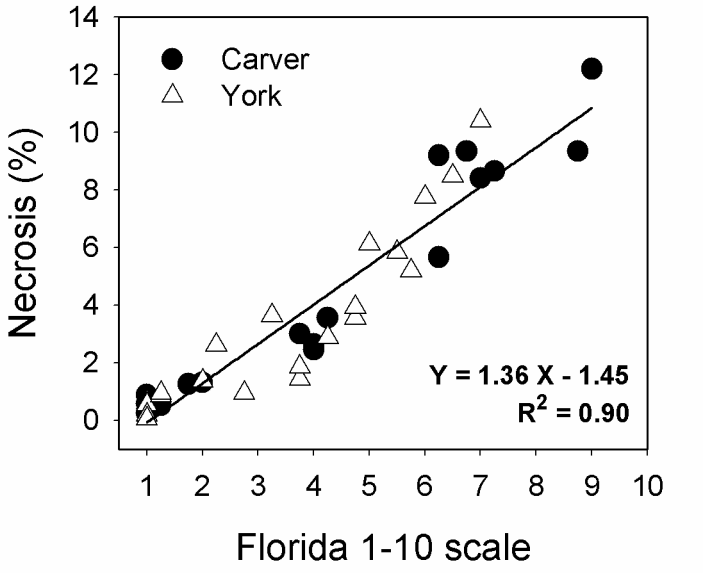

Figure 5-8. The relationship between percent necrosis and Florida 1-10 visual rating scale.

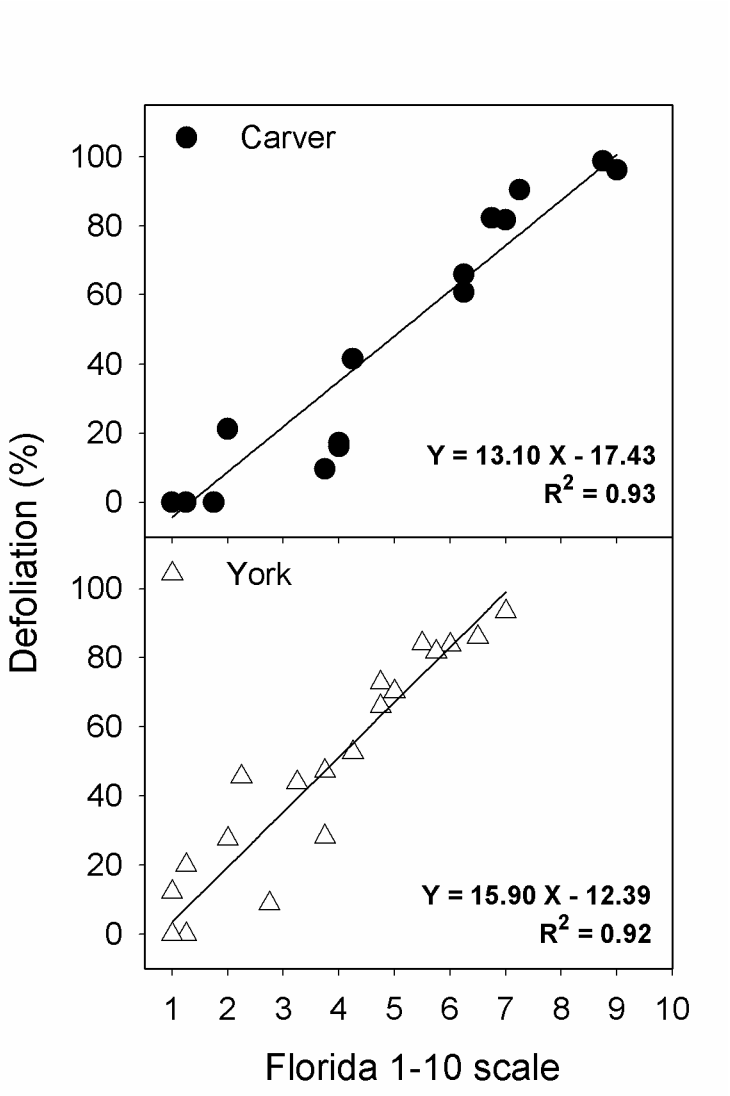

Figure 5-9. The relationship between defoliation and Florida 1-10 visual rating scale for the two peanut cultivars Carver and York.

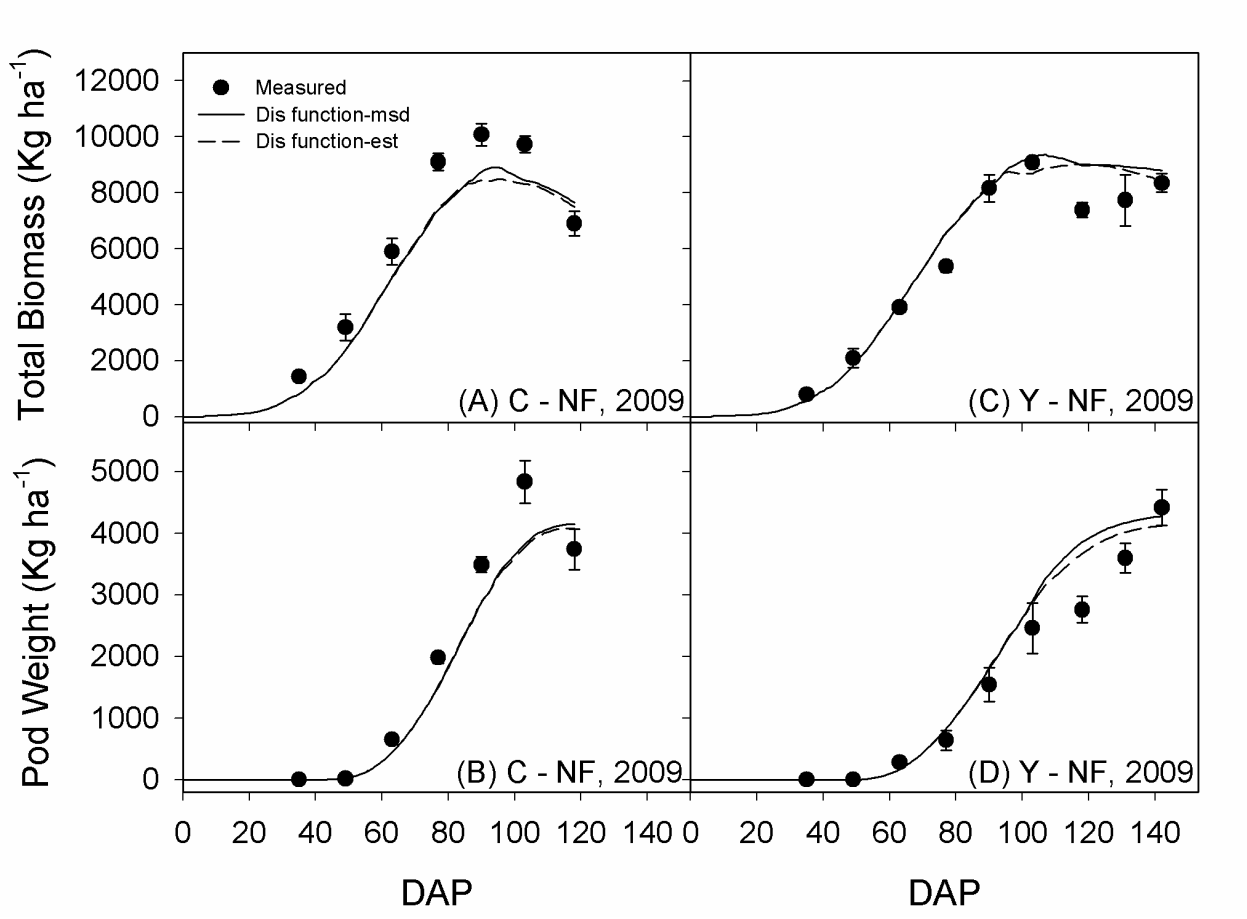

Figure 5-10. Simulated and measured total biomass and pod weight for peanut cultivars Carver (C) and York (Y) grown under no-fungicide application (NF) conditions during 2009. Symbols represents measured treatment means (*n*=4). Lines represents simulations with disease function derived from measured data (Dis function-msd) and estimated data from Florida 1-10 scale (Dis function-est). Vertical bars represent ± standard error of the mean.

CHAPTER 6
SUMMARY AND CONCLUSIONS

The overall goal of this study was to characterize LLS severity and progression and its impact on growth, yield and photosynthetic metabolism of peanut cultivars with differing levels of resistance to LLS. To that end, field experiments were conducted during the growing season of 2008 and 2009 at Citra, FL evaluating two peanut cultivars with more (York) and less (Carver) quantitative resistance to LLS grown under fungicide-sprayed and non-sprayed conditions. In addition, a simulated experiment was created based on field experiment data to predict the growth and yield reductions associated with LLS.

This study demonstrated that the more resistant cultivar York contributed to delayed disease progress resulting in slower development of canopy lesion area and reduced defoliation. Despite this, yield improvement over the less resistant cultivar, Carver, was marginal and only occurred during the second year of the study when LLS pressure was high. A diminishing effect of fungicide on the more resistant cultivar for pod yield was not observed, as yield gains associated with fungicide application were the same for both cultivars across both years of the study. So, foliar application of fungicides still played an important role in minimizing crop yield loss caused by LLS epidemics. These findings were attributed in part to a lack of improved physiological tolerance to LLS in York as shown by similar reductions in TCP in both cultivars despite reduced disease severity in York. These results indicate that combining resistance to disease progression with enhanced ability to sustain canopy photosynthetic capacity in the cultivar selection procedure could provide significant improvement in our efforts to improve peanut yields under diseased conditions.

This study also demonstrated variability in the leaf level photosynthetic response to LLS between cultivars with differing resistance levels to LLS. York, the cultivar with the higher level of resistance to LLS showed more photosynthetic impairment beyond the necrotic lesion area at a given disease severity compared to Carver, the cultivar with a poorer level of resistance. Possible mechanisms responsible for this greater photosynthetic impairment in York included a reduction in carboxylation velocity of Rubisco and/or reduction in chlorophyll content. These findings were attributed in part to a lack of photosynthetic tolerance to LLS in the more resistant cultivar York. Thus, reductions in photosynthetic rate in the virtual area were most likely due to a decline in protein content. This decline could be occurring uniformly throughout the non-necrotic leaf area in response to LLS infection rather than just the area around the necrotic lesions. These results have potential implications in our efforts for selecting improved cultivars and predicting growth and yield responses of new peanut cultivars to LLS. For example, combining visual disease ratings with physiological measures of the β parameter could result in the identification and selection of cultivars with slow disease progress (e.g., like York) and relatively low β parameters (e.g., like Carver), which could contribute to reduced yield loss due to LLS, especially under low fungicide input production.

In this study, the CROPGRO-Peanut model was successfully used to simulate the influence of foliar diseases (e.g. late leaf spot) on photosynthesis, growth, partitioning, and yield reductions in peanut cultivars with differing levels of resistance to LLS given inputs on canopy necrotic area and defoliation. Estimating defoliation from main stem nodes worked well, but obtaining the appropriate starting point (node 7 in Carver and

node 9 in York) for disease-induced defoliation is important. Significant relations between the Florida 1-10 visual rating scale and measured necrosis and defoliation can be used to simulate LLS-induced growth and yield reductions where detailed sampling on disease damage is not conducted. The adjustments made to the model code were relatively minor but resulted in improved predictions of the effect of LLS on growth and development of peanut. So, the next version of the CROPGRO model should include the changes to simulate effects of necrotic area directly on leaf photosynthetic traits. Use of a cultivar-specific β parameter was warranted as it resulted in improved simulations of growth and yield. The sensitivity of the model to percent necrosis (PDLA) and β parameter among different cultivars needs further investigation. Future model development should include an independent disease simulator which can predict LLS-induced necrosis and defoliation based on weather conditions. This would allow improved predictions of the impacts of foliar diseases like LLS on growth and yield, and hence result in reduced fungicide use and improved cultivar development without relying on the disease damage inputs from scouting.

APPENDIX A
DEFOLIATION, NECROSIS, DRY BIOMASS, AND CANOPY PHOTOSYNTHESIS VALUES FOR CARVER AND YORK

Table A-1. Calculated percent canopy defoliation (PCLA) and necrosis (PDLA) for peanut cultivars Carver and York grown under fungicide-sprayed (Fung) and non-sprayed (NF) conditions during 2008 across various DAP (days after planting) and DoY (day of year). PCLA values were calculated based on leaf weight loss (LW), main stem defoliation (MS), branch defoliation (Branch), and main stem defoliation excluding first six and eight nodes in Carver and York (MS + offset).

	Fungicide			PCLA (%)				
Cultivar	treatment	DAP	DoY	LW	MS	Branch	MS+offset	PDLA (%)
Carver	NF	35	176	0.0	0.0	0.0	0.0	0.00
		49	190	0.0	0.0	0.0	0.0	0.00
		64	205	0.0	13.6	0.0	0.0	0.25
		77	218	0.0	19.1	16.6	0.0	0.55
		91	232	0.0	23.2	39.8	0.0	1.26
		105	246	16.0	39.3	52.5	19.9	2.65
		119	260	82.2	90.6	89.3	87.9	9.33
		128	269	98.7	100.0	99.3	100.0	9.33
	Fung	35	176	0.0	0.0	0.0	0.0	0.00
		49	190	0.0	0.0	0.0	0.0	0.00
		64	205	0.0	11.3	0.0	0.0	0.37
		77	218	0.0	20.1	14.8	0.0	0.49
		91	232	0.0	24.4	42.9	1.7	0.88
		105	246	21.2	47.7	59.1	29.9	1.31
		119	260	41.4	65.4	73.6	54.2	3.55
		133	274	90.3	95.7	94.4	94.2	8.65
York	NF	35	176	0.0	0.0	0.0	0.0	0.00
		49	190	0.0	0.0	0.0	0.0	0.00
		64	205	0.0	16.3	0.0	0.0	0.36
		77	218	0.0	26.6	22.9	0.0	0.54
		91	232	12.1	42.7	52.5	13.6	0.62
		105	246	27.5	57.9	60.9	39.6	1.37
		119	260	43.7	65.0	76.3	49.2	3.63
		133	274	84.1	88.2	90.7	84.3	5.83
		147	288	93.4	98.7	98.0	98.2	10.4
	Fung	35	176	0.0	0.0	0.0	0.0	0.00
		49	190	0.0	0.0	0.0	0.0	0.00
		64	205	0.0	17.4	0.0	0.0	0.15
		77	218	0.0	22.9	31.0	0.0	0.50
		91	232	0.0	39.0	44.9	11.0	0.54
		105	246	19.9	54.7	63.4	35.4	0.86
		119	260	45.6	58.1	81.6	40.2	2.61
		133	274	66.0	75.9	82.9	66.0	3.55
		147	288	83.6	94.6	91.3	92.5	7.75

Table A-2. Calculated percent canopy defoliation (PCLA) and necrosis (PDLA) for peanut cultivars Carver and York grown under fungicide-sprayed (Fung) and non-sprayed (NF) conditions during 2009 across various DAP (days after planting) and DoY (day of year). PCLA values were calculated based on leaf weight loss (LW), main stem defoliation (MS), branch defoliation (Branch), and main stem defoliation excluding first six and eight nodes in Carver and York (MS + offset).

	Fungicide			PCLA (%)				
Cultivar	treatment	DAP	DoY	LW	MS	Branch	MS+offset	PDLA (%)
Carver	NF	35	182	0.0	0.0	0.0	0.0	0.00
		49	196	0.0	0.0	0.0	0.0	0.00
		63	210	0.0	18.1	0.0	0.0	0.31
		77	224	0.0	23.9	13.6	0.0	0.58
		90	237	9.6	43.2	41.3	22.1	3.00
		103	250	60.8	66.2	85.3	54.1	9.19
		118	265	96.1	92.7	96.7	90.3	12.20
	Fung	35	182	0.0	0.0	0.0	0.0	0.00
		49	196	0.0	0.0	0.0	0.0	0.00
		63	210	0.0	16.7	0.0	0.0	0.24
		77	224	0.0	22.8	14.5	0.0	0.25
		90	237	0.0	35.9	45.2	14.1	0.51
		103	250	17.1	41.9	53.1	21.6	2.45
		118	265	65.9	68.3	78.7	57.0	5.66
		125	272	81.6	87.8	93.9	84.1	8.40
York	NF	35	182	0.0	0.0	0.0	0.0	0.00
		49	196	0.0	0.0	0.0	0.0	0.00
		63	210	0.0	23.8	0.0	0.0	0.09
		77	224	0.0	34.7	18.5	0.0	0.18
		90	237	0.0	42.2	47.0	11.6	0.93
		103	250	28.2	59.1	73.4	40.4	1.44
		118	265	72.7	81.9	91.9	75.0	3.92
		131	278	81.6	86.6	91.9	81.7	5.19
		142	289	85.9	88.3	95.0	84.4	8.47
	Fung	35	182	0.0	0.0	0.0	0.0	0.00
		49	196	0.0	0.0	0.0	0.0	0.00
		63	210	0.0	24.4	0.0	0.0	0.12
		77	224	0.0	24.1	20.6	0.0	0.05
		90	237	0.0	42.7	61.0	16.8	0.05
		103	250	8.7	57.7	69.2	37.5	0.94
		118	265	47.3	65.5	80.4	50.6	1.86
		131	278	52.6	81.1	93.1	73.9	2.87
		142	289	70.1	85.5	93.6	80.4	6.11

Table A-3. Leaf, stem, pod, and total dry weight (DW) and leaf area index (LAI) mean values (n = 4) vs. days after planting (DAP) and day of year (DoY) for peanut cultivars Carver and York grown under fungicide-sprayed (Fung) and non-sprayed (NF) conditions during 2008.

Cultivar	Fungicide	DAP	DoY	LAI	Leaf DW	Stem DW	Pod DW	Total DW
Carver	NF	35	176	1.13	405	393	0	798
		49	190	2.72	1074	1318	5	2396
		64	205	5.28	2034	3113	506	5654
		77	218	4.84	2074	3434	1453	6960
		91	232	5.44	2235	4252	2515	9036
		105	246	4.00	1877	4229	4006	10189
		119	260	0.69	398	4088	4340	8897
		128	269	0.03	30	3144	4064	7274
	Fung	35	176	1.13	405	393	0	798
		49	190	2.72	1074	1318	5	2396
		64	205	4.31	1747	2637	359	4744
		77	218	4.72	1976	3161	1285	6422
		91	232	4.90	1979	3852	3085	8959
		105	246	3.34	1559	3771	3615	8995
		119	260	2.26	1159	3326	3571	8064
		133	274	0.95	191	3489	4442	8224
York	NF	35	176	0.58	222	171	0	393
		49	190	1.86	758	890	0	1649
		64	205	2.93	1150	1551	70	2770
		77	218	5.54	2315	3279	705	6299
		91	232	5.60	2035	3324	1041	6428
		105	246	3.60	1678	3433	1798	6971
		119	260	2.77	1300	3560	3517	8462
		133	274	2.15	369	2982	3283	6743
		147	288	0.28	153	2770	3249	6214
	Fung	35	176	0.58	222	171	0	393
		49	190	1.86	758	890	0	1649
		64	205	3.48	1345	1786	122	3252
		77	218	5.35	2102	2922	661	5685
		91	232	6.48	2455	4052	1760	8302
		105	246	4.71	1966	4228	1831	8100
		119	260	2.88	1336	3650	3592	8679
		133	274	2.70	835	2962	3409	7298
		147	288	0.80	402	3462	4029	7934

Table A-4. Leaf, stem, pod, and total dry weight (DW) and leaf area index (LAI) mean values (n = 4) vs. days after planting (DAP) and day of year (DoY) for peanut cultivars Carver and York grown under fungicide-sprayed (Fung) and non-sprayed (NF) conditions during 2009.

Cultivar	Fungicide	DAP	DoY	LAI	Leaf DW	Stem DW	Pod DW	Total DW
Carver	NF	35	182	1.81	661	768	0	1429
		49	196	2.50	1518	1660	12	3191
		63	210	4.28	2001	3250	651	5902
		77	224	5.44	2602	4504	1979	9096
		90	237	4.93	2352	4221	3487	10075
		103	250	1.72	1020	3868	4836	9725
		118	265	0.17	102	3057	3738	6897
	Fung	35	182	1.81	661	768	0	1429
		49	196	2.50	1518	1660	12	3191
		63	210	4.02	1885	2842	714	5442
		77	224	4.38	2096	3406	1926	7447
		90	237	4.02	2219	4353	3162	9750
		103	250	2.79	1839	3665	4800	10304
		118	265	0.98	756	3433	4572	8761
		125	272	0.58	407	3655	4691	8754
York	NF	35	182	1.20	417	383	0	800
		49	196	2.03	1016	1079	2	2097
		63	210	3.53	1551	2086	281	3918
		77	224	4.49	1950	2734	642	5382
		90	237	6.08	2672	3898	1545	8165
		103	250	3.84	1919	4702	2463	9084
		118	265	1.21	730	3900	2761	7391
		131	278	0.78	491	3645	3598	7734
		142	289	0.56	301	3632	4420	8352
	Fung	35	182	1.20	417	383	0	800
		49	196	2.03	1016	1079	2	2097
		63	210	3.64	1597	2270	162	4030
		77	224	3.99	1696	2521	733	5017
		90	237	5.88	2493	4384	1784	8701
		103	250	4.60	2276	4142	2597	9015
		118	265	2.17	1315	3818	3489	8622
		131	278	1.83	1181	3804	3848	8832
		142	289	1.16	673	4170	4991	9834

Table A-5. Mid-day total canopy photosynthesis (TCP) mean values (n = 2) vs. days after planting (DAP) and day of year (DoY) for peanut cultivars Carver and York grown under fungicide-sprayed (Fung) and non-sprayed (NF) conditions during 2008 and 2009.

		2008			2009		
Cultivar	Fungicide	DAP	DoY	TCP	DAP	DoY	TCP
Carver	NF	35	176	0.93	44	191	1.29
		49	190	1.29	56	203	1.76
		64	205	1.93	64	211	1.64
		78	219	1.66	78	225	1.78
		98	239	1.46	89	236	1.35
		107	248	1.06	103	250	0.46
		116	257	0.73	117	264	0.16
		128	269	0.05			
	Fung	35	176	0.93	44	191	1.29
		49	190	1.64	56	203	1.56
		64	205	1.72	64	211	1.6
		78	219	1.59	78	225	1.7
		98	239	1.3	89	236	1.48
		107	248	1.11	103	250	1.12
		116	257	0.92	117	264	0.46
		130	271	0.26			
York	NF	35	176	0.69	44	191	0.91
		49	190	1.06	56	203	1.29
		64	205	1.66	64	211	1.53
		78	219	1.87	78	225	1.85
		98	239	1.8	89	236	1.67
		107	248	1.35	103	250	1.29
		116	257	1.07	117	264	0.74
		130	271	0.38	132	279	0.5
		142	283	0.2	142	289	0.17
		147	288	0.2			
	Fung	35	176	0.69	44	191	0.91
		49	190	1.23	56	203	1.19
		64	205	1.65	64	211	1.47
		78	219	1.77	78	225	1.91
		98	239	1.6	89	236	1.62
		107	248	1.42	103	250	1.53
		116	257	1.32	117	264	1
		130	271	0.65	132	279	0.85
		142	283	0.34	142	289	0.35
		147	288	0.25			

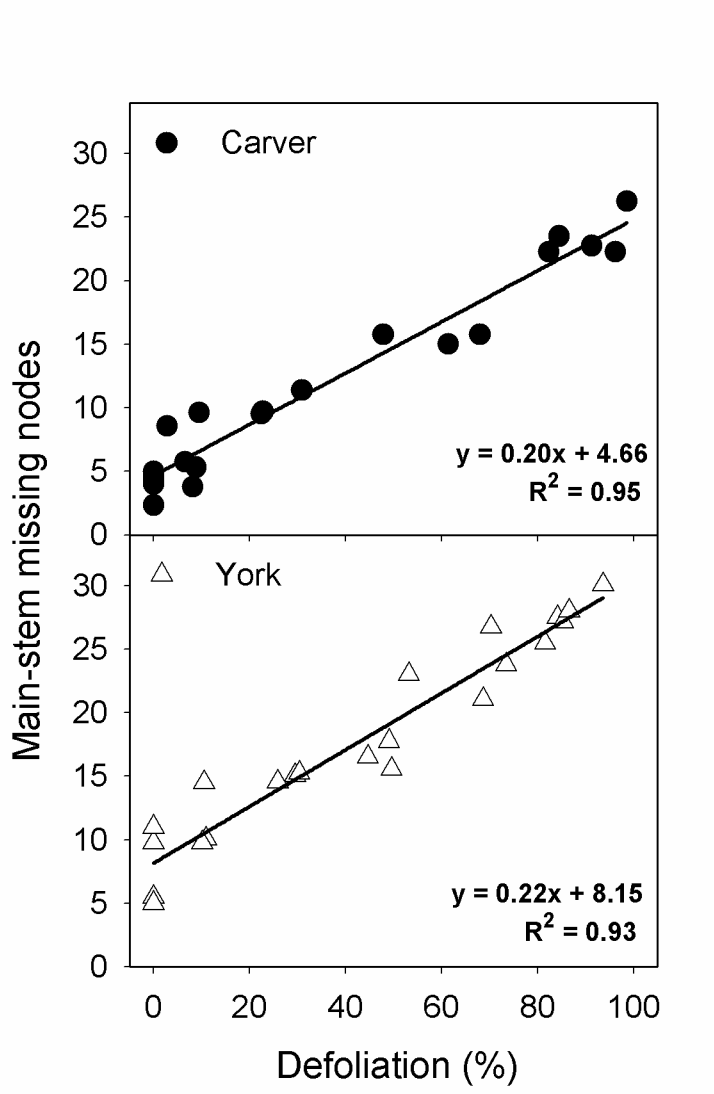

Figure A-1. Relationship between missing nodes on the main-stem and defoliation (based on leaf weight loss from peak weight) for the two peanut cultivars Carver and York.

APPENDIX B
LEAF SPOT RATING SCALES

Table B-1. Florida 1-10 rating scale (based on Chiteka et al., 1988).

Rating	Description
1	No disease
2	Very few lesions (none on upper canopy)
3	Few lesions (very few on upper canopy)
4	Some lesions with more on upper canopy and slight defoliation (≈5%)
5	Lesions noticeable even on upper canopy with noticeable defoliation (≈20%)
6	Lesions numerous and very evident on upper canopy with significant defoliation (≈50%)
7	Lesions numerous on upper canopy with much defoliation (≈75%)
8	Upper canopy covered with lesions with high defoliation (≈90%)
9	Very few leaves remaining and those covered with lesions (some plants completely defoliated)
10	Plants completely defoliated and killed by leaf spot

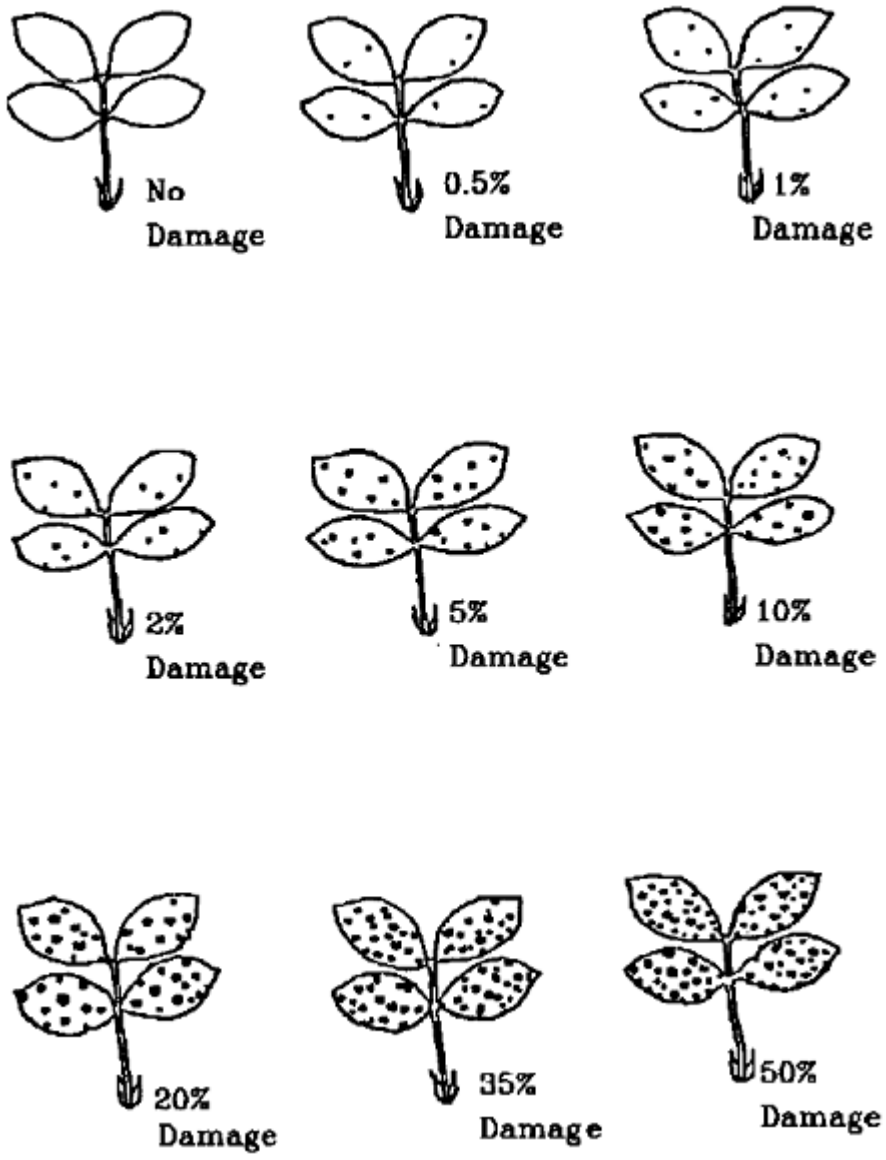

Figure B-1. ICRISAT diagrammatic scale to estimate percent leaflet necrosis (from Subrahmanyam et al., 1995).

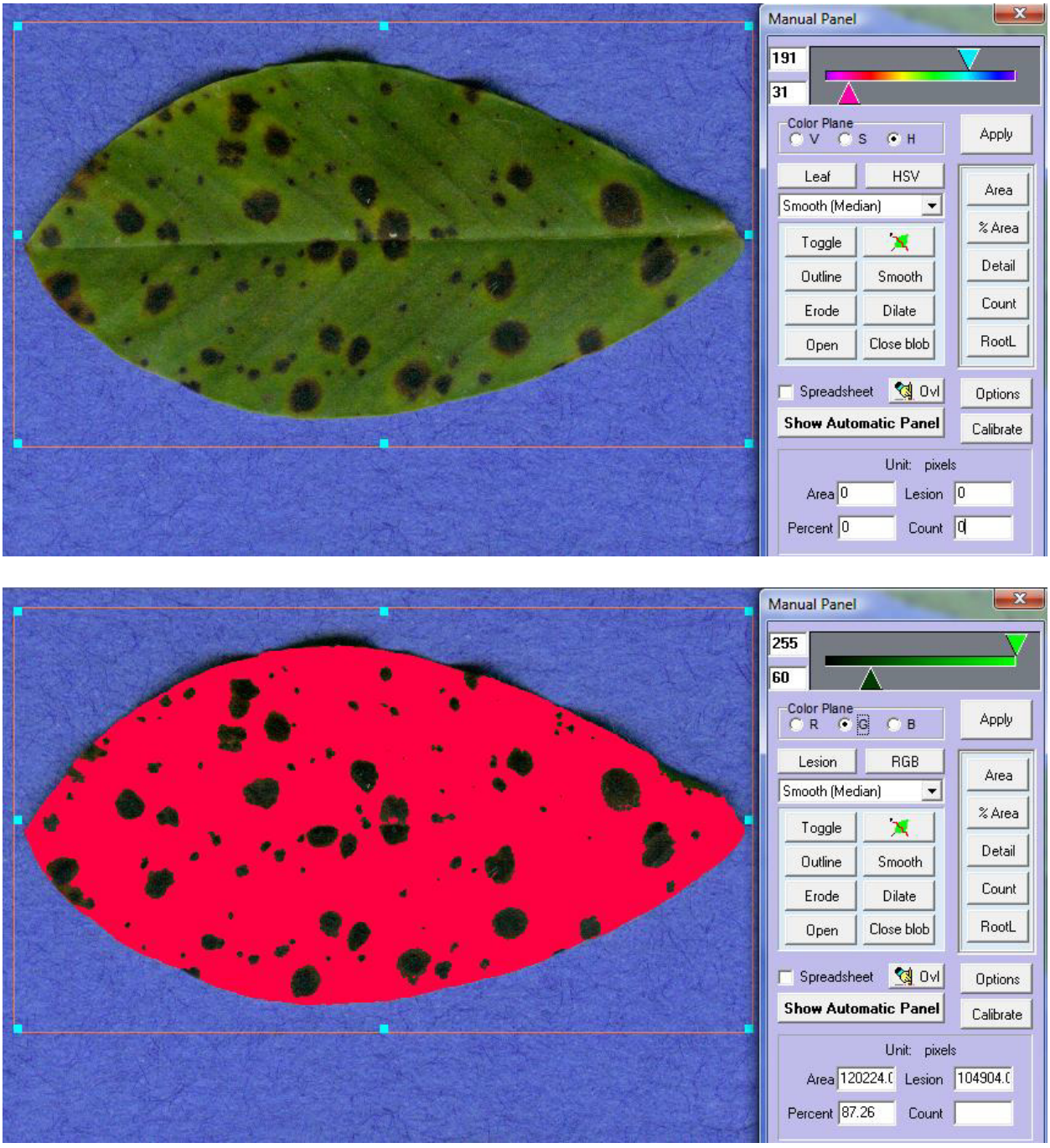

Figure B-2. Determination of percent necrotic leaf area using ASSESS ver 2.0 image analysis software. (A) scanned leaf showing necrotic spots, and (B) non-necrotic leaf area selected using the software. Percent necrotic area was determined by subtracting non-necrotic area from 100.

LIST OF REFERENCES

Abdou, Y. A-M., W.C. Gregory, and W.E. Cooper. 1974. Sources and nature of resistance to *Cercospora arachidicola* Hori and *Cercosporidium personatum* (Beack. And Curtis) Deighton in *Arachis* species. Peanut Sci. 1:6-11.

Adomou, M., P.V.V. Prasad, K.J. Boote, and J. Detongnon. 2005. Disease assessment methods and their use in simulating growth and yield of peanut crops affected by leafspot disease. Ann. Appl. Biol. 146:469-479.

Anderson, W.F., C.C. Holbrook, and T.B. Brenneman. 1993. Resistance to *Cercosporidium personatum* within peanut germplasm. Peanut Sci. 20:53-57.

Aquino, V.M., F.M. Shokes, D.W. Gorbet, and F.W. Nutter. 1995. Late leaf spot progression on peanut as affected by components of partial resistance. Plant Dis. 79:74-78.

Bancal, Marie-Odile, C. Robert, and B. Ney. 2007. Modelling wheat growth and yield losses from late epidemics of foliar diseases using loss of green leaf area per layer and pre-anthesis reserves. Ann. Bot. (London). 100:777-789.

Bassanezi, R.B., L. Amorim, A. Bergamin Filho, and R.D. Berger. 2002. Gas exchange and emission of chlorophyll fluorescence during the monocycle of rust, angular leaf spot and anthracnose on bean leaves as a function of their trophic characterization. J. Phytopathol. 150:37-47.

Bassanezi, R.B., L. Amorim, A. Bergamin Filho, B. Hau, and R.D. Berger. 2001. Accounting for photosynthetic efficiency of bean leaves with rust, angular leaf spot and anthracnose to assess crop damage. Plant Pathol. 50:443-452.

Bastiaans, L. 1991. Ratio between virtual and visual lesion size as a measure to describe reduction in leaf photosynthesis of rice due to leaf blast. Phytopathology 81:611-615.

Bastiaans, L. 1993. Effect of leaf blast on photosynthesis of rice. 1. Leaf photosynthesis. Neth. J. Pl. Path. 99:197-203.

Batchelor, W.D., J.W. Jones, K.J. Boote, and H.O. Pinnschmidt. 1993. Extending the use of crop models to study pest damage. Trans. ASAE 36:551-558.

Bergamin Filho, A., S.M.T.P.G. Carneiro, C.V. Godoy, L. Amorim, R.D. Berger, and B. Hau. 1997. Angular leaf spot of *Phaseolus* beans: Relationship between disease, healthy leaf area, and yield. Phytopathology 87:506-515.

Boote, K.J. 1999. Concepts of calibrating crop growth models. p. 179-200. *In* G. Hoogenboom et al. (ed.) DSSAT Version 3. A decision support system for agrotechnology transfer. Vol. 4. Univ. of Hawaii, Honolulu.

Boote, K.J., W.D. Batchelor, J.W. Jones, H. Pinnschmidt, and G. Bourgeois. 1993. Pest damage relations at the field level. p. 277-296. *In* F.W.T. Penning de Vries et al. (ed.) Systems approaches for agricultural development. Kluwer Acad. Publ., Dordrecht, the Netherlands.

Boote, K.J., J.M. Bennett, and J.W. Jones. 1983b. Canopy Photosynthesis: Apparent, Net, or Gross? *In* Advances in Photosynthesis Research. Proc. 6th Int. Congr. on Photosynthesis, Vrije Univ., Brussels, Belgium. 1-6 Aug. 1983. Dr. W. Junk Publ., The Hauge.

Boote, K.J., J.W. Jones, and J.M. Bennett. 1985. Factors influencing crop canopy CO_2 assimilation of soybean. p 780-785. *In* R. Shibles (ed.) Proc. World Soybean Research Conference III, 12-17 August 1984, Ames, IA. Westview Press, Boulder, CO.

Boote, K.J., J.W. Jones, and G. Hoogenboom. 1998a. Simulation of crop growth: CROPGRO model. p. 651-692. *In* R.M. Peart and R.B. Curry (ed.) Agricultural Systems Modeling and Simulation. Marcel Dekker, New York.

Boote, K.J., J.W. Jones, G. Hoogenboom, and N.B. Pickering. 1998b. The CROPGRO model for grain legumes. p. 99-128. *In* G.Y. Tsuji et al. (ed.) Understanding Options for Agricultural Production. Kluwer Academic Publishers, Dordrecht The Netherlands.

Boote, K. J., J. W. Jones, J. W. Mishoe, and R. D. Berger. 1983a. Coupling pests of crop growth simulators to predict yield reductions. Phytopathology 73: 1581-1587.

Boote, K. J., J. W. Jones, G. H. Smerage, C. S. Barfield, and R. D. Berger. 1980. Photosynthesis of peanut canopies as affected by leafspot and artificial defoliation. Agron. J. 72:247-252.

Bourgeois, G., and K. J. Boote. 1992. Leaflet and canopy photosynthesis of peanut affected by late leaf spot. Agron. J. 84:359-366.

Bourgeois, G., K.J. Boote, and R.D. Berger. 1991. Growth, development, yield, and seed quality of Florunner peanut affected by late leafspot. Peanut Sci. 18:137-143.

Branch, W.D. 2002. Registration of 'Georgia-01R' peanut. Crop Sci. 42:1750-1751.

Branch, W.D. 2006. Registration of 'Georgia-05E' peanut. Crop Sci. 46:2305.

Branch, W.D., and T. Brenneman. 2008. Registration of 'Georgia-07W' Peanut. J. Plant Registrations 2:88-91.

Butler, D.R., K.D.R. Wadia, and R.K. Reddy. 1995. Effects of humidity, leaf wetness, temperature and light on cinidial production by *Phaeoisariopsis personata* on groundnut. Plant Pathol. 44:662-674.

Cantonwine, E.G., A.K. Culbreath, C.C. Holbrook, and D.W. Gorbet. 2008. Disease progress of early leaf spot and components of resistance to *Cercospora arachidicola* and *Cercosporidium personatum* in runner-type peanut cultivars. Peanut Sci. 35:1-10.

Cantonwine, E.G., A.K. Culbreath, K.L. Stevenson, R.C. Kemerait, Jr., T.B. Brenneman, N.B. Smith, and B.G. Mullinix, Jr. 2006. Integrated disease management of leaf spot and spotted wilt of peanut. Plant Dis. 90:493-500.

Chiteka, Z.A., D.W. Gorbet, F.M. Shokes, T.A. Kucharek, and D.A. Knauft. 1988. Components of resistance to late leaf spot in peanut. I. Levels of variability – implications for selection. Peanut Sci. 15:25-30.

Cook, M. 1981. Susceptibility of peanut leaves to *Cercosporidium personatum*. Phytopathology 71:787-791.

Duncan, W.G., D.E. McCloud, R.L. McGraw, and K.J. Boote. 1978. Physiological aspects of peanut yield improvement. Crop Sci. 18:1015-1020.

Dwivedi, S.L., S. Pande, J.N. Rao, and S.N. Nigam. 2002. Components of resistance to late leaf spot and rust among interspecific derivatives and their significance in a foliar disease resistance breeding in groundnut (*Arachis hypogaea* L.). Euphytica 125:81-88.

Erickson, J.E., G.R. Stanosz, and E.L. Kruger. 2003. Photosynthetic consequences of Marssonina leaf spot differ between two poplar hybrids. New Phytol. 161:577-583.

FAO. 2011. FAOSTAT. Available at http://faostat.fao.org/default.aspx (verified 6 June 2011) FAO, Rome, Italy.

FAWN. 2011. Available at http://fawn.ifas.ufl.edu/ (verified 6 June, 2011). University of Florida, Gainesville, FL.

Farquhar, G.D., S. von caemmerer, and J.A. Berry. 1980. A biochemical model of photosynthetic CO_2 assimilation in leaves of C_3 species. Planta 149:78-90.

Gauch Jr., H.G., and G.B. Chase. 1974. Fitting the Gaussian curve to ecological data. Ecology 55:1377-1381.

Gorbet, D.W. 2006. Registration of ‘Carver’ peanut. Crop Sci. 46:2713-2714.

Gorbet, D.W. 2007. Registration of ‘Hull’ Peanut. J. Plant Registrations 1:125-126.

Gorbet, D.W., A. J. Norden, F.M. Shokes, and D. A. Knauft. 1987. Registration of ‘Southern Runner’ peanut. Crop Sci. 27:817.

Gorbet, D. W., and F.M. Shokes F. 2002a. Registration of ‘C-99R’ peanut. Crop Sci. 42:2207.

Gorbet, D. W., and F.M. Shokes F. 2002b. Registration of 'Florida MDR 98' peanut. Crop Sci. 42:2207-2208.

Gorbet, D.W. and Tillman, B.L. 2008. Registration of 'DP-1' peanut. J. Plant Registrations 2:200-204.

Gorbet, D.W., and B.L. Tillman. 2011. Registration of 'York' peanut. J. Plant Registrations 5:1-6.

Gregory, M.P., and W.C. Gregory. 1979. Exotic germplasm of *Arachis* L. interspecific hybrids. J. Hered. 70:185-193.

Guinn, G. and D.L. Brummett. 1993. Leaf age, decline in photosynthesis, and changes in abscisic acid, indole-3-acetic acid, and cytokinin in cotton leaves. Field Crops Res. 32:269-275.

Holbrook, C.C., and A.K. Culbreath A. 2007. Registration of 'Tifrunner' Peanut. J. Plant Registrations 1:124.

Holbrook, C.C., and A.K. Culbreath. 2008. Registration of 'Georganic' peanut. J. Plant Registrations 2:17.

Holbrook, C.C., P. Timper, A.K. Culbreath, and C.K. Kvien. 2008. Registration of 'Tifguard' peanut. J. Plant Registrations 2:92-94.

Hoogenboom, G., J.W. Jones, P.W. Wilkens, C.H. Porter, K.J. Boote, L.A. Hunt, U. Singh, J.L. Lizaso, J.W. White, O. Uryasev, F.S. Royce, R. Ogoshi, A.J. Gijsman, and G.Y. Tsuji. 2009. Decision Support System for Agrotechnology Transfer (DSSAT) Version 4.5 [CD-ROM] University of Hawaii, Honolulu, Hawaii.

Inskeep, W.P., and P.R. Bloom. 1985. Extinction coefficients of chlorophyll a and b in N,N-dimethylformamide and 80% acetone. Plant Physiol. 77:483-485.

Jackson, L.F. 1981. Distribution and severity of peanut leafspot in Florida. Phytopathology 71:324-328.

Jackson, C.R., and D.K. Bell. 1969. Diseases of peanut (groundnut) caused by fungi. University of Georgia. Coll. Agric. Exp. Stn. Res. Bull. 56:1-5.

Jacobi, J.C., and P.A. Backman. 1995. AU-Pnuts Advisory II: Modification of the rule-based leaf spot advisory system for a partially resistant peanut cultivar. Plant Dis. 79:672-676.

Jacobi, J.C., P.A. Backman, D.P. Davis, and P.M. Brannen. 1995. AU-Pnuts Advisory I: Deveolopment of a rule based system for scheduling peanut leaf spot fungicide application. Plant Dis. 79:666-671.

Jenkins, W. 1938. Two fungi causing leafspot of peanuts. J. Agric. Res. 56:317-332

Jesus Jr., W.C., F.X.R. do Vale, R.R. Coelho, B. Hau, L. Zambolim, L.C. Costa, and A. Bergamin Filho. 2001. Effects of angular leaf spot and rust on yield loss of *Phaseolus vulgaris*. Phytopathology 91:1045-1053.

Jones, J.W., G. Hoogenboom, C.H. Porter, K.J. Boote, W.D. Batchelor, L.A. Hunt, P.W. Wilkens, U. Singh, A.J. Gijsman, and J.T. Ritchie. 2003. The DSSAT cropping systems model. Europ. J. Agron. 18:235-265.

Kemerait, B., T. Brenneman, and A. Culbreath. 2010. Peanut disease update. p 57-80. *In* J. P. Beasley, Jr. (ed.) 2010 Peanut Update. University of Georgia Ext., Georgia.

Knauft, D.A., and D.W. Gorbet. 1990. Variability in growth characteristics and leafspot resistance parameters of peanut lines. Crop Sci. 30:169-175.

Knauft, D.A., A.J. Norden, D.W. Gorbet, and F.G. Martin. 1987. Stability of market quality factors in peanut (*Arachis hypogaea* L.). Soil and Crop Science Society of Florida Proceedings 46:72-74.

Krapovickas, A., and W.C. Gregory. 1994. Taxonomy of the genus *Arachis* (Leguminosae). Bonplandia 8:1-186.

Kucharek, T. 2005. Extension plant pathology report no. 12, Disease control program for peanuts. Plant Pathology department. University of Florida/IFAS, Gainesville, FL.

Kumudini, S., C.V. Godoy, B. Kennedy, E. Prior, J. Omielau, H.R. Boerma, and D. Hershman. 2010. Role of host-plant resistance development stage on leaf photosynthetic competence of soybean rust infected leaves. Crop Sci. 50:2533-2542.

Kumudini, S., E. Prior, J. Omielan, and M. Tollenaar. 2008. Impact of *Phakopsora pachyrhizi* infection on soybean leaf photosynthesis and radiation absorption. Crop Sci. 48: 2343-2350.

Leal-Bertioli, S.C. D.M., M.C.D. Farias, P.I.T. Silva, P.M. Guimaraes, A.C.M. Brasileiro, D.J. Bertioli, and A.C.G. D. Araujo. 2010. Ultrastructure of the initial interaction of Puccinia arachidis and Cercosporidium personatum with leaves of Arachis hypogaea and Arachis stenosperma. J Phytopathol. 158:792-796.

Lin, L., A.S. Hedayat, B. Sinha, and M. Yang. 2002. Statistical methods in assessing agreement: models, issues and tools. J. Amer. Stat. Assoc. 97:257-270.

Long, S.P., and C.J. Bernacchi. 2003. Gas exchange measurements, what can they tell us about the underlying limitations to photosynthesis? Procedures and sources of error. J. Expt. Bot. 54:2393-2401.

Lopes, D.B., and R.D. Berger. 2001. The effects of rust and anthracnose on the photosynthetic competence of disease bean leaves. Phytopathology 91:212-220.

McDonald, D., P. Subrahmanyam, R.W. Gibbons, and D.H. Smith. 1985. Early and late leaf spots of groundnut. Info. Bull. no. 21. International Crops Research Institute for the Semi-Arid Tropics. Patancheru, Andhra Pradesh, India.

Mims, C.W., E.S. Luttrell, and S.C. Alderman. 1988. Ultrastructure of haustorium of the peanut late leaf spot fungus *Cercosporidium personatum*. Can. J. Bot. 67:1198-1202.

Monfort, W.S., A.K. Culbreath, K.L. Stevenson, T.B. Brenneman, D.W. Gorbet, and S.C. Phatak. 2004. Effects of reduced tillage, resistant cultivars, and reduced fungicide inputs on progress of early leaf spot of peanut (*Arachis hypogaea*). Plant Dis. 88:858-864.

Moriondo, M., S. Orlandini, A. Giuntoli, and M. Bindi. 2005. The effect of downy and powdery mildew on grapevine (*Vitis vinifera* L.) leaf gas exchange. J. Phytopathology 153: 350-357.

Morton, B.R. 2007. Poor field emergence of late-maturing peanut cultivars (*Arachis hypogaea* L.) derived from PI-203396.Dissertation. University of Florida, Gainesville, FL.

Mossler, M. and M.J. Aerts. 2007. Florida crop/pest management profiles: peanuts. CIR 1260. Florida Coop. Ext. Serv., University of Florida, Gainesville, FL.

Naab, J.B., P. Singh, K.J. Boote, J.W. Jones, and K.O. Marfo. 2004. Using the CROPGRO-Peanut model to quantify yield gaps of peanut in the Guinean savanna zone of Ghana. Agron. J. 96:1231-1242.

Nogues, S., L. Cotxarrera, L. Alegre, and M.I. Trillas. 2002. Limitations to photosynthesis in tomato leaves induced by Fusarium wilt. New Phytol. 154:461-470.

Nutter Jr., F.W., and F.M. Shokes. 1995. Management of foliar diseases caused by fungi. p. 65-74. *In* H.A. Melouk and F.M. Shokes (eds.) Peanut Health Management. American Phytopathological Society, St. Paul, MN.

Perfect, S.E., and J.R. Green. 2001. Infection structures of biotrophic and hemibiotrophic plant pathogens. Molecular Pl. Pathology 2:101-108.

Phakamas, N., A. Patanothai, S. Jogloy, K. Pannangpetch, and H. Hoogenboom. 2008. Physiological determinants of pod yield of peanut lines. Crop Sci. 48:2351-2360.

Pinnachmidt, H.O., W.D. Batchedlor, and P.S. Teng. 1995. Simulations of multiple species pest damage on rice. Agric. Syst. 48:193-222.

Pinnachmidt, H.O., Y. Luo, and P.S. Teng. 1994. Methodology for quantifying rice yield effects of blast. p 318-408. *In* R.S. Ziegler et al (ed.) Rice blast diseases. CAB International, Wallingford, Oxon, UK.

Pixley, K.V., K.J. Boote, F.M. Shokes, and D.W. Gorbet. 1990a. Growth and partitioning characteristics of four peanut genotypes differing in resistance to late leafspot. Crop Sci. 30:796-804.

Pixley, K.V., K.J. Boote, F.M. Shokes, and D.W. Gorbet. 1990b. Disease progression and leaf area dynamics of four peanut genotypes differing in resistance to late leafspot. Crop Sci. 30:789-796.

R Development Core Team. 2008. R: A language and environment for statistical computing. R foundation for statistical computing, Vienna, Austria.

Robert, C., Marie-Odile Bancal, C. Lannou, and B. Ney. 2006. Quantification of the effects of *Septoria tritici* blotch on wheat leaf gas exchange with respect to lesion age, leaf number, and leaf nitrogen status. J. Exp. Bot. 57:225-234.

Robert, C., Marie-Odile Bancal, B. Ney, and C. Lannou. 2005. Wheat leaf photosynthesis loss due to leaf rust, with respect to lesion development and leaf nitrogen status. New Phytol. 165:227-241.

Robert, C., Marie-Odile Bancal, P. Nicolas, C. Lannou, and B. Ney. 2004. Analysis and modellling of effects of leaf rust and *Septoria tritici* blotch on wheat growth. J. Exp. Bot. 55:1079-1094.

Roloff, I., H. Scherm, and M. W. Iersel. 2004. Photosynthesis of blueberry as affected by septoria leaf spot and abiotic leaf damage. Plant Dis. 88:397-401.

SAS Institute. 2009. The SAS system for Windows. Release 9.2. SAS Inst., Cary, NC.

Shanner, G., and R.E. Finney. 1977. The effect of nitrogen fertilization on the expression of slow-mildewing resistance in knox wheat. Phytopathology 67:1051-1056.

Shew, B.B., M.K. Beute, and J.C. Wynne. 1988. Effect of temperature and relative humidity on expression of resistance to *Cercosporidium personatum* in peanut. Phytopathology 78:493-498.

Shokes, F.M., and A.K. Culbreath. 1997. Early and late leaf spots. p. 17-20. *In* N. Kokalis-Burelle, D.M. Porter, R. Rodriguez-Kabana, D.H. Smith, and P. subrahmanyam (eds.) Compendium of Peanut Diseases, 2nd Ed. APS Press, St. Paul, MN.

Shokes, F.M., D.W. Gorbet, and L. Jackson. 1983. Control of early and late leaf spot on two peanut cultivars. Peanut Sci. 10:17-21.

Shokes, F.M., D.W. Gorbet, and G.E. Sanden. 1982. Effect of planting date and date of spray initiation on control of peanut leaf spots in Florida. Plant Dis. 66:574-575.

Smith, D.H., and R.H. Littrell. 1980. Management of peanut foliar diseases with fungicides. Plant Dis. 64:356-361.

Smith, D.H., G.D.C. Paucer, and F.M. Shokes. 1992. Cercosporidium and cercospora leaf spots of peanut (Groundnut). p. 285-304. *In* U.S. Singh, A.N. Mukhopadhyay, J. Kumar, and H.S. Chaube (eds.) Plant Diseases of International Importance. Prentice Hall Inc., Englewood Cliffs, New Jersey.

Shtienberg, D. 1992. Effect of foliar disease on gas exchange process: a comparative study. Phytopathology 82:760-765.

Stalker, H.T. 1997. Peanut (*Arachis hypogaea* L.). Field Crops Res. 53:205-217.

Subrahmanyam, P., D. McDonald, F. Waliyar, L.J. Reddy, S.N. Nigam, R.W. Gibbons, V. Ramanatha Rao, A.K. Singh, S. Pande, P.M. Reddy, and P.V. Subba Rao. 1995. Screening methods and sources of resistance to rust and late leaf spot of groundnut. ICRISAT Information Bulletin No. 47. ICRISAT, Patancheru, Andhra Pradesh, India.

Teng, P.S., W.D. Batchelor, H.O. Pinnschmidt, and G.G. Wilkerson. 1998. Simulation of pest effects on crops using coupled pest-crop models: the potential for decision support. p 221-266. *In* G.Y. Tsuji et al (ed.) Understanding options for agricultural production. Kluwer Academic Publishers, Dordrecht The Netherlands.

Tillman, B., D. Gorbet, M. Gomillion, J. McKinney, G. Person, and B. Thomas. 2008. Peanut variety performance in Florida 2004-2007. SS AGR 311. Florida Coop. Ext. Serv., Univ. of Florida, Gainesville, FL.

Tillman, B.L., and H.T. Stalker. 2009. Peanut. p 287-317. *In* J. Vollmann and I. Rajcan (ed.) Oil Crops, Handbook of plant breeding 4. Springer Science.

Timsina, J., K.J. Boote, and S. Duffield. 2007. Evaluating the CROPGRO soybean model for predicting impacts of insect defoliation and depodding. Agron. J. 99:148-157.

USDA NASS. 2011. Crop Acreage and Value: Peanut. Available at http://www.nass.usda.gov (verified 6 June 2011). Washington, D.C.

Williams, J.H., and K.J. Boote. 1995. Physiology and modeling- predicting the "unpredictable legume". p 301-353. *In* H.E. Pattee and H.T. Stalker (ed.) Advances in peanut science. Am. Peanut Res. And Edu. Soc., Stillwater, Ok.

Williams, E.J., and J.S. Drexler. 1981. A non-destructive method for determining peanut pod maturity. Peanut Sci. 8:134-141.

Willmott, C.J. 1981. On the validation of models. Physical Geography 2:184-194.

Willmott, C.J. 1982. Some comments on the evaluation of model performance. Bull. Am. Meteorol. Soc. 63:1309-1313.

Woodward, J.E., T.B. Brenneman, R.C. Kemerait, Jr., A.K. Culbreath, and N.B. Smith. 2010. Management of peanut diseases with reduced input fungicide programs in fields with varying levels of disease risk. Crop Prot. 29:222-229.

Woodward, J.E., T.B. Brenneman, R.C. Kemerait, Jr., N.B. Smith, A.K. Culbreath, and K.L. Stevenson. 2008. Use of resistant cultivars and reduced fungicide programs to manage peanut disease in irrigated and nonirrigated fields. Plant Dis. 92:896-902.

Wright, D.L., B. Tillman, E. Jowers, J. Marois, J.A. Ferrell, T. Katsvairo, and E.B. Whitty. 2006. Management and cultural practices for peanuts. SS AGR 74. Florida Coop. Ext. Serv., Univ. of Florida, Gainesville, FL.

Wynne, J.C., M.K. Beute, and S.N. Nigam. 1991. Breeding for disease resistance in peanut. Annu. Rev. Phytopathol. 29:279-303.

Yin, X., J. Goudriaan, E.A. Lantinga, J. Vos, and H.J. Spiertz. 2003. A flexible sigmoid function of determinate growth. Ann. Bot. (London) 91:361-371.

Zhang, S., S. Lu, X. Xu, H. Korpelainen, and C. Li. 2009. Changes in antioxidant enzyme activities and isozyme profiles in leaves of male and female *Populus cathayana* infected with *Melampsora larici-populina*. Tree Physiol. 30:116-128.

BIOGRAPHICAL SKETCH

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