EFFECT OF SOUTHERN ROOT-KNOT NEMATODE (*MELOIDOGYNE INCOGNITA*) ON COTTON GROWTH, YIELD, AND FIBER QUALITY

by

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(Under the Direction of Robert C. Kemerait, Jr.)

ABSTRACT

Southern root-knot nematode [*Meloidogyne incognita* (Kofoid & White) Chitwood] is a major parasite on cotton (*Gossypium hirsutum L.*). In this study, greenhouse experiments were conducted to evaluate the effect of *M. incognita* on cotton growth and physiological variables related to fiber development. Field experiments were conducted to evaluate the effect of different nematode management strategies on cotton growth, yield, and fiber quality. In greenhouse trials, most plant growth and physiological variables showed significant reduction with *M. incognita* infection. In field trials, Telone II significantly reduced *M. incognita* populations and increased cotton growth compared with Temik 15G and seed treatments. Cotton lint yield was improved by Telone II in most field trials except in two out of four fields in 2009. Most fiber quality properties were not consistently or significantly affected by different nematicide treatments across fields and years. Nematicides performed similarly in different nematode management zones.

INDEX WORDS: Cotton, Fiber quality, Growth, *Meloidogyne incognita*, Site-specific management, Southern root-knot nematode, Yield

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CHAPTER 1

INTRODUCTION AND LITERATURE REVIEW

Cotton (*Gossypium hirsutum L.*), is an oilseed and fiber crop. It is grown in more than seventy countries and is the single most important fiber crop worldwide. No crop competes with it in the potential of value-added processing (Basra, 1999). Cotton is also one of the oldest cultivated crops. It has been associated with human activity since before recorded history.

The U.S. is the third-largest producer of cotton in the world. Cotton production in the U.S. has increased 66% in the past 40 years (Mitchell, 2009). In recent years, the U.S. has produced about 20% of the world's annual supply (Mitchell, 2009).

Root knot nematodes (*Meloidogyne* spp.) occur worldwide and attack a diversity of crops. They can cause considerable losses of yield and affect the quality of the products, either by directly damaging plants (Kirkpatrick and Sasser, 1984) or by predisposing them to infection by fungal and bacterial pathogens (Powell, 1971).

The southern root-knot nematode [*Meloidogyne incognita* (Kofoid & White) Chitwood] is found in all cotton production regions in the U.S. and is the most widely distributed nematode parasite of economic importance to the crop (Thomas and Kirkpatrick, 2001). It is considered the major yield-limiting plant-parasitic nematode across the U.S. Cotton Belt; approximately twice as much yield loss is attributed to southern root-knot nematodes than to all other nematode parasites of the crop (Koenning et al., 2004). The estimated yield loss of cotton caused by *M. incognita* in the U.S. was 2.4% in 2007, which was greater than for any other cotton disease.

This damage resulted in a loss of more than 106,000,000 kg of lint (Cotton Disease Loss Committee, 2008). In Georgia in 2007, the southern root-knot nematode caused an estimated 6% reduction in yield resulting in a loss of 25,000,000 kg of lint (Cotton Disease Loss Committee, 2008).

Damage caused by plant-parasitic nematodes is distinct from other plant diseases because there are no unique symptoms on the above-ground portions of the plants. Nematodes are often unevenly distributed in the soil; therefore the symptoms associated with damage from nematodes may occur in irregular patches in the field (Beltwide Cotton Committee, 2003). These patches can be either small and limited in number, or large and widely distributed. Damaged plants may exhibit symptoms ranging from mild to severe stunting, depending on disease severity, and a reduced rate of development. Foliage may also show symptoms of nutritional deficiency (Kirkpatrick et al., 1995). In the most severe cases, plants may die before maturation. The nematodes also interact with the Fusarium wilt pathogen (*F. oxysporum* f.sp. *vasinfectum*), which leads to wilting and brown discoloration or necrosis of the vascular tissue of the lower stem (Beltwide Cotton Committee, 2003).

Below-ground symptoms caused by *M. incognita*s on cotton can be much more diagnostic than above-ground symptoms. Visible galls or "knots" often appear on cotton roots (Bridge and Page, 1980). Swellings of the infected root tissue can be found on the cotton tap root and the lateral roots; however, the galls on cotton may not be as easy to observe as those on vegetables such as tomatoes. Galls are easier to detect if cotton plants are carefully dug (not pulled) from the soil. Also, the fine lateral roots need to be carefully handled when rinsed with water to remove soil (Beltwide Cotton Committee, 2003).

The tap root and its lateral roots are of vital importance to the cotton plant. A few galls on these roots can disrupt the normal flow of water and nutrients to the leaves and developing bolls, which can significantly reduce the cotton yield (Bird and Loveys, 1975; Kirkpatrick et al., 1991; McClure, 1977).

It has been shown that plant growth can be reduced significantly by heavy infection with *M. incognita* (Bird, 1970). Above-ground symptoms of *M. incognita* infection are nondescript, but include suppressed plant growth (stunting), nutritional deficiency (chlorosis), and temporary wilting during the heat of the day (Beltwide Cotton Committee, 2003). Most damage to the cotton plant caused by *M. incognita* results from physiological changes caused by nematode feeding on root tissues. Reduced plant growth and leaf expansion as a result of infection by *M. incognita* have been documented (Kirkpatrick et al., 1995). Infection by *M. incognita* has also reduced the number and size of cotton bolls and plant dry weight (Walker et al., 1998). Anatomically, *M. incognita* affects cotton roots by disrupting the xylem as well as changing root epidermis and cortical tissues in response to giant-cell development and gall formation.

Research has shown that infection by *M. incognita* can reduce photosynthetic rates in different plants. Within 2 days of *M. incognita* infection, differences between photosynthetic rates in control and test tomato plants occurred (Loveys and Bird, 1973). This seemed to be a physiological response rather than a morphological one. However, after 22 days, morphological differences had become important in determining the CO_2-fixing capacity of the whole plant. It has been suggested that during early stages of infection, the decreased photosynthesis was highly significant when expressed on the basis of fresh weight, leaf area, or total chlorophyll content (Loveys and Bird, 1973). This physiological response may have resulted from a reduced supply of root-derived photosynthesis-regulating factors. For example, it was shown that both

cytokinins and gibberellins in tomato root tissue and xylem exudates were decreased in plants infected with *M. incognita* compared with control plants (Brueske and Bergeson, 1972). During late stages of the infection, the reduced rate of photosynthesis was at least partly due to the smaller size of infected plants (Loveys and Bird, 1973).

It has also been discovered that *M. incognita* infection suppressed plant growth and reduced yield, chlorophyll content, photosynthetic rate, and nutrient concentrations in Henbane (*Hyoscyamus niger*). The greatest reduction in all characters was observed when plants were inoculated with the highest nematode population (Haseeb et al., 1990).

Previous research has focused on the management of *M. incognita* to improve cotton yield. Very little data has documented the impact of *M. incognita* on cotton fiber quality. However, cotton fiber quality is a very important issue to the growers and to the textile industry. Fiber quality is assessed using a set of measurements that describe a sample of fibers extracted from a bale of cotton (Bradow and Davidonis, 2000). These measurements include length, uniformity, strength, micronaire, color grade, trash, leaf grade, preparation, and extraneous matter (USDA, 2001). They are compared with a set of standards from the U.S. Department of Agriculture (USDA) and are used to determine price premiums and discounts.

In recent years, cotton lint quality has become increasingly important because of the requirements by textile mills for high fiber quality, the use of high-volume instrument (HVI) testing, the occurrence of discounts due to unfavorable fiber characteristics, and depressed cotton markets (Silvertooth, 1999). Georgia is the third-largest cotton producer in the U.S. In the 1990s and early 21st century, cotton produced in Georgia had fiber quality second only to that produced in California, but now certain fiber quality parameters have not only fallen below levels found in

western U.S cotton, but also below the average fiber quality of other southern and southeastern states (Bradow and Davidonis, 2000).

Cotton plants are sensitive to stress and there are many factors that could influence fiber quality. As has been described in the literature, nutrients, moisture, insecticides and environmental factors all have impact on one or more fiber properties because they interrupt one or more processes of fiber physiological development (Pettigrew, 2001; Reddy et al., 2004; Roberts et al., 2005; Shimishi and Masani, 1971; Snipes and Baskin, 1994).

There are four general phases in cotton fiber development: 1) initiation, 2) elongation, 3) thickening, and 4) desiccation (maturation). Fiber length is determined in the elongation stage, which occurs about 21 days after flowering. During this stage, a thin cell wall of carbohydrate polymers is deposited allowing the fiber to elongate (DeLanghe, 1986). Water pressure inside the developing fiber has an influence on fiber elongation by regulating the deposition of carbohydrate polymers (Bradow and Davidonis, 2000). Therefore, if stresses occur during this stage (which could be associated with water stress or potassium deficiency, for example), fiber length can be reduced (Silvertooth, 1999). To some degree, fiber strength and uniformity can also be influenced by these same stresses (Bradow and Davidonis, 2000).

The thickening process of cotton fibers may overlap with elongation to some extent. During this stage, carbohydrates produced through photosynthesis are deposited on the interior walls of the fiber, which increases the value of micronaire (Silvertooth, 1999). If this development is stopped prematurely, finer fibers will occur and the micronaire value will decrease. New bolls that are set on the plant usually have some of the greatest demand for carbohydrates. These bolls draw carbohydrates from the older bolls, which prevents the development of high micronaire fibers on older bolls (Silvertooth, 1999).

The maturation of fiber occurs after the boll has opened and the metabolically inactive fibers dry. There is no quality measurement directly related to the maturation process. The fiber quality within a boll is at its utmost on the day of boll opening (Bradow and Davidonis, 2000). Therefore harvesting should be as close to physiological maturity as possible to enhance the quality of the crop produced.

Environmental factors and management factors can significantly alter cotton fiber quality. For example, reduced light (cloudy) conditions result in the production of weaker fibers with reduced micronaire (Pettigrew, 2001). Early defoliation can also reduce micronaire in cotton (Snipes and Baskin, 1994).

Biological factors such as plant-pathogenic fungi and insect pests have also been studied. Cotton root rot, caused by the soilborne fungus *Phymatotrichum omnivorum*, can reduce fiber length and fineness significantly (Mulrean, 1984). It has also been well documented that by feeding on cotton bolls, stink bugs can decrease fiber quality as indicated by a reduction in almost all HVI-measured variables (Roberts et al., 2005). *M. incognita*, a root-feeding plant parasite, is another factor that could potentially affect fiber quality. *M. incognita* draws nutrients and water from cotton plants and exacerbates moisture, nutrient, or other stresses and also fungal infection, which could indirectly damage fiber development.

In previous studies, several fiber properties were documented to be affected by moisture and nutrient stress, and reduced or delayed cotton growth. In other studies, it has been shown that similar stresses could be caused by *M. incognita*.

Fiber length and staple can be influenced by several factors including cotton variety, temperature, water stress, nutrient deficiencies, and ginning practices (Bradow and Davidonis, 2000). Water relationships and irrigation practices have been studied primarily in relation to

yield. One study conducted in the early 1980s indicated that fiber length was not impacted until water stress was such that yields were limited to less than 706.1 kg/ha (Grimes and Yamada, 1982). The interaction between *M. incognita* and water stress in cotton has also been studied. The results indicated that in susceptible cultivars, infection by *M. incognita* may decrease the movement of water from roots to leaves. The decrease in root flux caused by nematodes is equal to that induced by severe water deficit stress (Kirkpatrick et al., 1991). Other studies have demonstrated that water stress early in the bloom period had a less negative impact on fiber length than water stress late in the bloom period (Hearn, 1976; Marani and Amirav, 1971; Shimishi and Masani, 1971). Sensitivity of fiber elongation to severe water stress is apparently due to the physiological and mechanical processes of cell expansion (Hearn, 1994). As *M. incognita* infection becomes increasingly severe later in the season, the resulting water stress in the cotton plant could lead to a reduction in fiber length.

The possible impact of *M. incognita* on fiber strength is not as direct as the impact on fiber length. Firstly, fiber strength has a positive relationship with canopy sunlight absorption. There is evidence that *M. incognita* can reduce plant leaf area, e.g., in tomato (Loveys and Bird, 1973). Therefore by reducing cotton leaf area, *M. incognita* may affect fiber strength. Secondly, studies have indicated that heat accumulation during the flowering period can also affect fiber strength. Fiber strength was greatest in bolls that developed from flowers produced during the first 4 to 6 weeks of flowering. Flowers that opened during the latter 2 weeks of the flowering period produced bolls with the lowest fiber strength (Jones and Wells, 1997). Therefore, *M. incognita* may reduce fiber strength by delaying cotton development and flowering.

Authors in a previous study have suggested several factors that could influence fiber micronaire. Significant differences in micronaire among commercially available varieties have

been examined. A fiber thickens as cellulose is deposited inside the fiber cell, resulting in increased micronaire values. If cotton photosynthesis is affected by *M. incognita*, as confirmed in tomato plants (Loveys and Bird, 1973), the lack of carbohydrate production may lead to a lower micronaire value. However, a micronaire value that is too high can also result in loss of profit. It is important for cotton to produce sufficient bolls later in the season to either compete for carbohydrates or to produce lower micronaire fiber, which can be blended with the higher micronaire fiber to reduce overall readings (Silvertooth, 1999). Therefore, the reduced growth and boll setting in cotton caused by *M. incognita* (Kirkpatrick et al., 1995; Walker et al., 1998) may affect the blended micronaire value.

As discussed previously, water stress can be caused by *M. incognita* (Kirkpatrick et al., 1991). In other studies, it has also been shown that water deficiency in cotton can affect some cotton fiber properties. Moisture stress later in the season was found to reduce fiber length and micronaire (Marani and Amirav, 1970). A reduction in fiber length due to water stress was also observed in a later study. Although other characters were not consistently affected, the distribution of bolls on cotton was consistently and significantly affected by irrigation (Pettigrew 2004).

Studies on the effect of *M. incognita* on nutrient concentrations in plants showed that a change in concentration of nutrients is likely one of the first effects of the nematode on host physiology. These changes in nutrient concentration alter host metabolism and contribute directly or indirectly to the chlorosis and premature leaf abscission in soybean (Melakeberhan et al., 1987). This is also true according to another study in soybean. The uptake of nitrogen, phosphorus, and calcium was affected by *M. incognita* (Carneiro et al., 2002). Many nutrients have effects on fiber quality. For example, leaf nitrogen during the boll maturation period had

significant positive correlations with fiber length and negative correlations with micronaire (Reddy et al., 2004). Potassium is also a very important element in cotton and has positive correlations with fiber length, micronaire, fiber strength, and fiber length-uniformity ratio (Cassman et al., 1990).

Approaches to effective nematode control include crop rotation, field sanitation, cover crops, varietal resistance, and nematicides. Nematode population densities can be reduced by selecting rotation crops that are not hosts to *M. incognita*, or by planting resistant cultivars (Rich and Kinloch, 2005). To date, no *M. incognita*-resistant cultivar is commercially available. Several cotton cultivars have been reported to have some level of tolerance to the Fusarium wilt/*M. incognita* disease complex. However, these cultivars do not show significant resistance to the nematodes, although some are resistant to Fusarium wilt (Beltwide Cotton Committee, 2003). Biological approaches for nematode control have been studied but have not been thoroughly explored in cotton production systems (Starr et al., 2007).

Chemical nematicides are widely used to control *M. incognita*. Three nematicide management strategies are currently widely used in the U.S. (Koenning et al., 2004), including the application of aldicarb at a rate of 0.8-1.2 kg/ha in the planting furrow; pre-plant soil fumigation using either 1,3-dichloropropene or metam-sodium (Starr et al., 2007); and the supplemental use of either aldicarb applied as a side-dress during the first third of the season or a foliar application of the carbamate oxamyl (Lawrence and Mclean, 2000, 2002). Out of all these strategies, at-planting application of aldicarb is perhaps the most universal nematicide strategy in the U.S. Aldicarb is applied on 20 to 30% of the cotton hectarage each year (Koenning, et al., 2004). In severely infested fields, nematicides can reduce the nematode population by more than 50% (Beltwide Cotton Committee, 2003). Using chemical nematicides can provide cotton with a

zone of protected soil in which roots can develop for 4 to 6 weeks with reduced damage from nematodes. By protecting the crop during early development, yield losses can be reduced substantially even though nematodes may penetrate the roots during the latter part of the season (Beltwide Cotton Committee, 2003). Seed treatments that include nematicides have shown some promising results in protecting emerging roots from nematode infection (Monfort et al., 2006), but may not be sufficient in fields with high nematode population densities.

In Georgia, commonly used commercial practices are the application of the soil fumigant Telone II (1,3 – dichloropropene) before planting and application of granular nematicides such as Temik 15G (aldicarb). However, the cost of nematicides may cause growers to use them sparingly in their production system (Ortiz et al., 2008). The seed treatments AVICTA Complete Cotton [AVICTA (abamectin), Cruiser (thiomethoxam) and Dynasty CST (azoxystrobin, fludioxonil and mefenoxam)] and AERIS Seed-Applied System (imidacloprid and thiodicarb) were both reported to have nematicidal effects, and therefore have the potential of being incorporated into *M. incognita* management strategies.

Like other soil-borne plant pathogens, *M. incognita* populations are present in the soil in an aggregated pattern and the damage they cause often appears as patches in the field. This aggregated distribution is affected primarily by the variability of soil properties. It has been shown that out of 26 edaphic components including soil texture, acidity, pH, and others, 50% of the variability in nematode density was related to high levels of clay, organic matter, low copper concentration, and small changes in percent soil moisture (Noe and Barker, 1985).

To offer a more cost-effective tactic for use of nematicides on cotton, a management zone strategy targeting areas with damaging nematode populations for treatment with nematicides has been studied. The management zone strategy is based on the fact that *M. incognita* populations

are present in the soil in aggregated patterns due to the variability of soil texture in a field, as *M. incognita* prefers coarse-textured, sandy soils. By using site-specific management (SSM) for nematicides, higher, more expensive rates of nematicides can be applied selectively to zones that potentially have higher nematode populations. A lower rate, or less expensive nematicides, can be targeted for low nematode risk zones. Hopefully, the efficacy of control of nematodes can be improved and expenses can be reduced.

In 2001, a strong correlation between Columbia lance nematode (*Hoplolaimus columbus*) densities and sand content was observed from electrical conductivity data that was used to map within-field spatial distribution of soil texture (Khalilian et al., 2001). This indicated promise for application of SSM against Columbia lance nematodes. In recent years, SSM has also been studied with *M. incognita* in cotton fields. The data indicated that edaphic features can be used as indirect indicators of high-risk, nematode-prone areas (Ortiz et al, 2006). In 2007, it was found that different nematicide rates had significantly different effects on cotton yields within different risk zones (Ortiz et al., 2008). These findings supported the strategy of variable-rate nematicide applications based on *M. incognita* risk management zones. The goal of using SSM in this study was to determine whether cotton fiber quality may also respond differently to nematicide applications in different management zones.

The objectives of the research presented in this thesis were to: (1) Determine the extent to which *M. incognita* causes physiological damage in cotton, based upon levels of *M. incognita* inoculum density and cotton varieties. (2) Examine the influence of *M. incognita* on cotton growth, yield, and fiber quality and the effect of the SSM strategy for managing cotton yield and fiber quality. And (3) determine the effects of *M. incognita* on growth, yield, fiber quality, and

economics of altered fiber quality as related to choice of variety. The methods and results will be presented in three different chapters.

In *Chapter 2*, the stress in cotton caused by *M. incognita* was quantified in three greenhouse trials. Different nematode levels were created by infesting soil with different quantities of *M. incognita* eggs. Several physiological variables presumed to be related to cotton fiber development were measured. This portion of the thesis research was conducted because of the difficulties in measuring such characters in field studies, and because it was easier to manipulate the nematode populations in the greenhouse.

In *Chapter 3*, the methods and results from three field trials in the 2008 and again in 2009 seasons are presented. For these trials, the field sites were delineated into two risk management zones based on several soil properties, as southern *M. incognita* maybe present in a field in a certain pattern with preference for coarse-textured, sandy soils (Noe and Barker, 1985; Starr et al., 1993; Koenning et al., 2004; Monfort et al., 2007). Some soil properties have been shown to be related to the presence or absence of *M. incognita*. These properties include fertility (Noe and Barker, 1985), pH (Melakeberhan et al., 2004), and moisture (Wheeler et al., 1991). Different nematicide and seed treatments were applied in the two risk management zones. Plant growth, root damage, nematode populations and reproduction, cotton yield, and fiber quality were measured throughout the growing seasons and after harvest. Differences in fiber properties among treatments and between zones are demonstrated in this chapter, as well as the correlation of nematode populations and disease severity with fiber properties. Principal component analysis was used to determine the factors which contribute most to the alteration of fiber properties.

In *Chapter 4,* field studies from two additional field sites are presented. At these two field sites, the site-specific management strategy was not applied. Treatments included fumigant,

granular in-furrow and seed treatment nematicides, and two cotton varieties with different levels of *M. incognita* resistance. Cotton growth, root damage, nematode population and reproduction, cotton yield, and fiber quality were measured in these fields. The results as related to lint yield and fiber properties. The relationships between *M. incognita* population density, cotton yield, and fiber quality are also discussed. The economic value of fiber quality with different nematicides, different risk management zones, and different cotton varieties is analyzed and discussed in this chapter.

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CHAPTER 2

EFFECT OF SOUTHERN ROOT KNOT NEMATODE (*MELOIDOGYNE INCOGNITA*) ON COTTON GROWTH AND PHYSIOLOGY IN THE GREENHOUSE[1]

[1] Lu, P., Kemerait, R. C., Jr., and Scherm, H. To be submitted to *Journal of Nematology*.

2.1 Abstract

To quantify the stress in cotton (*Gossypium hirsutum L.*) caused by southern root knot nematode [*Meloidogine incognita* (Kofoid & White) Chitwood], three greenhouse trials were conducted in 2008 and 2009. Soil planted to cotton was infested with two population densities of eggs of *M. incognita*. Cotton growth and several physiological variables thought to be related to cotton fiber development were examined. FiberMax 960BR and Stoneville 5599BR varieties were used in all three trials; breeding lines PD94042 and 120B1R1 were used in trials 2 and 3. In all three trials vegetative and reproductive growth, chlorophyll content, leaf expansion, plant weight, and photosynthetic rate were significantly reduced by *M. incognita* infection. Stoneville 5599BR had higher levels of tolerance and resistance to *M. incognita* infection compared with FiberMax 960BR, as documented by root gall ratings and nematode egg counts. Breeding line 120R1B1 had significant resistance to *M. incognita* in greenhouse trials.

Keywords: Cotton breeding lines, *Meloidogyne incognita*, Photosynthetic rate, Physiology, Southern root knot nematode

2.2 Introduction

Cotton is an oilseed and fiber crop. It is grown in more than seventy countries and is the single most important fiber crop worldwide. No crop competes with it in the potential of value-added processing (Basra, 1999). Cotton is also one of the oldest cultivated crops. It has been associated with human activity since before recorded history.

The U.S. is the third-largest producer of cotton in the world. Cotton production in the U.S. has increased 66% in the past 40 years. In recent years, the U.S. has produced about 20 percent of the world's annual supply (Mitchell, 2009).

Root knot nematodes occur worldwide and attack a diversity of crops. They cause considerable losses of yield and affect the quality of the products, either by directly damaging plants (Kirkpatrick and Sasser, 1984), or by predisposing them to infection by fungal and bacterial pathogens (Powell, 1971).

The southern root knot nematode [*Meloidogyne incognita* (Kofoid & White) Chitwood] is found in all cotton production regions in the U.S. and is the most widely distributed nematode parasite of economic importance to the crop (Thomas and Kirkpatrick, 2001). It is considered the major yield-limiting plant-parasitic nematode across the U.S. Cotton Belt; approximately twice as much yield loss is attributed to *M. incognita* than to all other nematode parasites of the crop (Koenning et al., 2004). The estimated yield loss of cotton caused by *M. incognita* in the U.S. was 2.4% in 2007, which was greater than for any other cotton disease. This damage resulted in a loss of more than 106,000,000 kg of lint (Cotton Disease Loss Committee, 2008). In Georgia in 2007, *M. incognita* caused an estimated 6% reduction in yield resulting in a loss of 25,000,000 kg of lint (Cotton Disease Loss Committee, 2008).

Damage caused by plant-parasitic nematodes is distinct from other plant diseases because there are no unique symptoms on the above-ground portions of the plants. Nematodes are often unevenly distributed in the soil; therefore the symptoms associated with damage from nematodes may occur in irregular patches in the field (Beltwide Cotton Committee, 2003). These patches can be either small and limited in number, or large and widely distributed. Damaged plants may exhibit symptoms ranging from mild to severe stunting, depending on disease severity, and a reduced rate of development. Foliage may also show symptoms of nutrient deficiency (Kirkpatrick et al. 1995). In the most severe cases, plants may die before maturation. The nematodes also interact with the Fusarium wilt pathogen (*Fusarium oxysporum* f.sp. *vasinfectum*), which leads to wilting and brown discoloration or necrosis of the vascular tissue of the lower stem (Beltwide Cotton Committee, 2003).

Below-ground symptoms caused by *M. incognita* on cotton can be much more diagnostic than above-ground symptoms. Visible galls or "knots" often appear on cotton roots (Bridge and Page, 1980). Swellings of the infected root tissue can be found on the cotton tap root and the lateral roots; however, the galls on cotton may not be as easy to observe as those on vegetables such as tomatoes. Galls are easier to detect if cotton plants are carefully dug (not pulled) from the soil. Also, the fine lateral roots need to be carefully handled when rinsed with water to remove soil (Beltwide Cotton Committee, 2003).

The tap root and its lateral roots are of vital importance to the cotton plant. A few galls on these roots can disrupt the normal flow of water and nutrients to the leaves and developing bolls, which can significantly reduce cotton yield (Bird and Loveys, 1975; Kirkpatrick et al., 1991; McClure, 1977).

It has been shown that plant growth can be significantly reduced by a heavy infection with *M. incognita* (Bird, 1970). Above-ground symptoms of *M. incognita* infection are nondescript, but include suppressed plant growth (stunting), nutritional deficiency (chlorosis), and temporary wilting during the heat of the day (Beltwide Cotton Committee, 2003). Most damage to the cotton plant caused by *M. incognita* results from physiological changes caused by nematodes feeding on root tissues. Reduced plant growth and leaf expansion as a result of infection by *M. incognita* have been documented (Kirkpatrick et al., 1995). Infection by *M. incognita* has also reduced the number and size of cotton bolls and plant dry weight (Walker et al., 1998). Anatomically, *M. incognita* affects cotton roots by disrupting the xylem as well as changing root epidermis and cortical tissues in response to giant-cell development and gall formation.

Although *M. incognita*-resistant cotton cultivars are not commercially available, there are cotton varieties that have partial resistance or tolerance, such as certain Stoneville varieties. Tolerance implies that the cultivar can yield well in the presence of the nematode, but does not necessarily suppress the reproduction of the nematode. Partial resistance indicates some reduction in nematode reproduction occurs on a cultivar (Wheeler et al., 2009). FiberMax varieties, although reported to be highly susceptible to damage from root knot nematode, are thought to produce a superior fiber quality and outrank other cultivars in fiber length and strength. FiberMax is reported to have fewer gin discounts for quality, which can bring more value to the growers.

Research has shown that infection by *M. incognita* can reduce photosynthetic rates in different plants. Within 2 days of *M. incognita* infection, differences between photosynthetic rates in control and test tomato plants occurred (Loveys and Bird, 1973). This seemed to be a

physiological response rather than a morphological one. However, after 22 days, morphological differences had become important in determining the CO_2-fixing capacity of the whole plant. It has been suggested that during early stages of infection, the decreased photosynthesis was highly significant when expressed on the basis of fresh weight, leaf area, or total chlorophyll content (Loveys and Bird, 1973). This physiological response may have resulted from a reduced supply of root-derived photosynthesis-regulating factors. For example, both cytokinins and gibberellins in tomato root tissue and xylem exudates were decreased in plants infected with *M. incognita* compared with control plants (Brueske and Bergeson, 1972). During late stages of the infection, the reduced rate of photosynthesis was at least partly due to the smaller size of infected plants (Loveys and Bird, 1973).

It has also been discovered that *M. incognita* infection suppressed plant growth, reduced yield, chlorophyll content, photosynthetic rate, and nutrient concentrations in Henbane (*Hyoscyamus niger*). The greatest reduction in these variables was observed when plants were inoculated with the highest nematode population (Haseeb et al., 1990).

Studies on the effect of *M. incognita* on nutrient concentrations in plants documented that a change in concentration of the nutrients in the plant is likely one of the first effects of the nematode on host physiology. These changes in nutrient concentration alter host metabolism and contribute directly or indirectly to the chlorosis and premature leaf abscission in soybean (Melakeberhan et al., 1987). This is also true according to another study in soybean. The uptake of nitrogen, phosphorus, and calcium was affected by *M. incognita* (Carneiro et al., 2002).

The objective of this chapter was to (1) further study damage of *M. incognita* in cotton to determine the degree to which growth and physiological variables were both affected negatively

by nematode infection; and (2) examine whether the damage caused by *M. incognita* was similar in different cotton cultivars that had different levels of resistance or tolerance.

2.3 Materials and Methods

Three greenhouse trials were conducted in Athens, GA, to document the effect of *M. incognita* on cotton growth and several physiological variables thought to be related to fiber development, including chlorophyll content, chlorophyll fluorescence, and photosynthetic rate. The three trials were conducted in spring 2008, fall 2008, and spring 2009. Each trial lasted 75 to 80 days from planting to destructive sampling.

Two cotton cultivars were used in the first trial, including FiberMax 960 BR, which is susceptible to *M. incognita* and Stoneville 5599 BR, which was developed from partially resistant cultivar ST LA887, and is considered to have both tolerance (Barfield, 2003) and partial resistance (Phipps and Eisenback, 2005) to *M. incognita*. Tolerance implies that the cultivar can yield well in the presence of the nematode, but does not necessarily suppress the reproduction of the nematode. Partial resistance indicates that some reduction in nematode reproduction occurs on a cultivar (Wheeler et al., 2009). FiberMax cultivars, though reported to be highly susceptible to damage from root knot nematodes, are thought to produce a superior fiber quality and outrank other cultivars in fiber length and strength. FiberMax is reported to have fewer gin discounts for quality, which can bring more value to the growers.

In trial 2 and trial 3, two cotton breeding lines were also used in addition to the two varieties included in trial 1. The two breeding lines were from a breeding program at the University of Georgia. One was PD94042, an upland cotton line with high yield and fiber quality (May, 1999) but no documented resistance to *M. incognita*. The other one was an unregistered

line 120B1R1, which was a cross between PD94042 and M-120 RNR, with PD94042 as the recurrent parent in a back-cross sequence. Breeding line M-120 RNR was derived from germplasm line Auburn 634 RNR, which has exceptionally high resistance to *M. incognita* and Fusarium wilt (Shepherd, 1982).

All cultivars were planted in clay plots with a diameter of 15.2 cm, and were placed on the same bench in the greenhouse. The medium used for greenhouse trials was a mixed soil, including 87.6% sand, 8.4% silt and 4% clay. At 10 days after planting, all plants were inoculated with *M. incognita* eggs in three different population densities. The eggs used for inoculation were extracted from *M. incognita*-infected roots of eggplants, which were cultured in the greenhouse previously. The three nematode densities that were applied included 0, 6,000 and 20,000 eggs per pot.

The experimental design used for the greenhouse trials was a split-plot design, with egg density being the main plot and cultivar being the sub-plot. In trials 2 and 3, the two breeding lines were tested as a separate set from the two commercial varieties, and the data were analyzed separately. There were 11 replications for trial 1 and 10 replications for trials 2 and 3. The plants were watered twice a day and fertilized once a week. The fertilizer contained 20% nitrogen, 20% phosphorus and 20% potassium. Half the normal amount of fertilizer, which was 30 mL fertilizer mixed with 18.9 L (5 gallons) water, was applied, as symptoms caused by nematodes are best shown in plants under stressed conditions. A final volume of 250 mL diluted fertilizer was applied to each plant. A mixture of the insecticides Merit (imidacloprid) and Enstar (s-kinoprene) was sprayed every week to control whiteflies. Each insecticide was applied at 0.65 mL/L (1/2 teaspoon/gallon) of water. For mite control, several materials were rotated every week, including Avid (abamectin) at 1.3 mL/L (1 teaspoon/gal), Floramite (bifenazate) at 1.3 mL/L (1

teaspoon/gal), and a mix of Orthene (acephate) 1.3 mL/L (1 teaspoon/gal), Telstar (bifenthrin) 1.3 ml/L (1 teaspoon/gal), and Enstar (s-kinoprene) 0.65 mL/L (1/2 teaspoon/gal).

Several measurements were collected weekly to record plant growth, including shoot heights, number of nodes, and height-to-node ratios. Shoot height was measured from the surface of the soil to the terminal bud. The number of nodes was determined by counting all nodes on the main stem except the cotyledonary nodes as long as the leaf associated with the node was bigger than a U.S. quarter coin. Height-to-node ratio is calculated as the shoot height divided by number of nodes. *M. incognita* infection could potentially reduce the elongation of plants (stunting); therefore height-to-node ratio can be used as an indicator of the stress in cotton caused by nematodes. Increased height-to-node ratios could indicate that lower stress occurred in plants. The chlorophyll content was measured using a Minolta SPAD-502 chlorophyll meter (Spectrum Technologies, Plainfield, IL), on the uppermost fully expanded leaf several times and the average number was recorded. The chlorophyll content was measured in the uppermost fully expanded leaves because these are usually the most active leaves on the plants. Also it is the easiest way to identify leaves with the same age in different plants. Chlorophyll fluorescence was measured in the evenings after the plants had been in the dark for at least 30 min. A pulse-amplitude modulation fluorometer (Heinz Walz GmbH, Effeltrich, Germany) was used to measure chlorophyll fluorescence on the uppermost fully expanded leaf.

All three variables, which were measured weekly, were transformed into areas under the variable progress curves using the trapezoidal method, which is the most commonly used method for estimating the area under the disease progress curve (AUDPC) (Madden et al., 2007). The formula used to calculate areas under progress curve in this study was the same as the formula for area under disease progress curves, which is $\mathrm{AUDPC} = \sum_{i=1}^{n-1}\left(\frac{y_i + y_{i+1}}{2}\right)\left(t_{i+1} - t_i\right)$. Here, t is

time in days after inoculation for each measurement, *y* is the reading of the variable that was measured, and *n* is the number of readings. The areas under the progress curve from each treatment were then analyzed using ANOVA and LSD tests in SAS PROC GLM (SAS Institute, Cary, NC).

At 75 to 80 days after planting, all cotton plants were destructively sampled for further measurements. Immediately prior to destructive sampling, photosynthetic rate was measured, using a CIRAS-1 portable photosynthesis measuring system (PP Systems, Amesbury, MA). Along with photosynthetic rate, stomatal conductance, leaf temperature, and sub-stomatal CO_2 concentration were also measured by CIRAS-1 at the same time. This measurement was obtained on the uppermost fully expanded leaf. After collecting the photosynthetic rate measurement, the plant tops were cut from the soil line, and the following measurements were taken: (1) Plant weight. The fresh weight of plant tops was measured immediately after destructive sampling. After all other measurements for the tops were completed; shoots were packed into paper bags and dried in an oven at 60˚C for 3 days. Thereafter, dry weight was also measured. (2) Leaf area. Total leaf area was measured by stripping leaves from the plants and feeding them through a LI-3100 Area Meter (LI-COR Biosciences, Lincoln, NE) (3) Number and weight of bolls. The number of cotton squares and bolls was collected and counted together as the number of bolls and their fresh weight and dry weight were also measured together as boll fresh weight and dry weight. (4) Root fresh weight. Cotton roots were washed thoroughly from soil and the fresh weight was then measured. Dry weight was not measured as this would interfere with further analysis for nematode damage. (5) Root gall rating. The galling severity for each root system was rated on a 0 to 5 scale for the greenhouse trials: 0 = no galling, 1 = trace infection with a few small galls, 2 = galling evident on <25% of the roots, 3 = 25 to 50%, 4 = 50 to 75% and 5

= >75% of the roots galled (Kinloch, 1990). (6) Egg counts. As a final processing step, nematode eggs produced on cotton roots were extracted with 0.625% NaOCl for 3 min (Hussey and Barker, 1973) and counted microscopically.

2.4 Results

2.4.1 *Meloidogyne incognita* infection and reproduction

In most cases, inoculation with *M. incognita* resulted in a significant increase in root gall ratings and egg counts per gram of root compared with the untreated control. However, the difference between mid and high nematode inoculation levels was usually not significant (Tables 2.1 and 2.2).

There was a significant variety effect on gall ratings and egg counts. FiberMax 960BR had significantly higher gall ratings and egg counts than Stoneville 5599BR in all three trials (Table 2.1). In most cases, FiberMax 960BR had approximately twice the gall rating and egg counts as Stoneville 5599BR. In trial 2, there were interactions between nematode population and variety in these measurements. The difference between plants grown in non-infested soil and plants grown in infested soil was more significant in FiberMax 960BR than in Stoneville 5599BR. The resistant breeding line 120R1B1 had root gall ratings and root necrosis ratings that were approximately half of the same ratings in breeding line PD94042 (Table 2.2). The egg counts per gram of root in 120R1B1 were only 1/14 of that observed in PD94042 in trial 2 and about 1/22 of that in PD94042 in trial 3.

2.4.2 Areas under the progress curves

Stunted growth is a common symptom caused by *M. incognita* in many crops. This also proved to be true in the greenhouse tests in this study. The area under the height-to-node ratio curve was significantly reduced with increased nematode populations, especially with the higher inoculation level (Tables 2.3 and 2.4). The area under the chlorophyll content progress curve was also reduced by *M. incognita* inoculation. The high level of nematode infestation typically resulted in significantly less chlorophyll content, but not in trial 2. *M. incognita* infestation generally had much less effect on the areas under the chlorophyll fluorescence progress curve. Although there was a numerical reduction in the chlorophyll fluorescence of commercial varieties, *M. incognita* infection only significantly reduced chlorophyll fluorescence in trial 3.

Comparing the two cotton varieties, Stoneville 5599BR consistently had a higher area under the height-to-node ratio progress curve than FiberMax 960BR, but had a smaller area under the chlorophyll content progress curve than FiberMax 960BR. The difference between varieties in area under the chlorophyll fluorescence progress curve was usually not significant, and was not consistent in different trials. In the two breeding lines, areas under the height-to-node ratio, chlorophyll content, and chlorophyll fluorescence progress curves were all higher in PD94042 in one trial, but the reverse was true in the other trial. There were no interactions between varieties and nematode inoculation levels or between breeding lines and nematode inoculation levels for any of these variables.

2.4.3 Cotton biomass

Cotton biomass, measured as shoot fresh weight, shoot dry weight, root weight, and total leaf area at 75 to 80 days after inoculation, showed significant reduction with *M. incognita*

inoculation in one or more trials (Tables 2.5 and 2.6). For the two commercial varieties, the reduction of biomass with the high level of nematode inoculation was significantly greater than the reduction of biomass with the mid-level of nematode inoculation, with the exception of root weight in trial 3. There were no statistical interactions between varieties and nematode inoculation levels on any of these variables, which indicated that the damage level caused in the two varieties by *M. incognita* was similar. In the two breeding lines, root weight was only significantly reduced in PD94042 in trial 3 (Table 2.6). Dry weight was significantly reduced in both breeding lines in both trials when comparing plants grown in inoculated with non-inoculated soils. Leaf area was also significantly reduced except for 120R1B1 in trial 3.

Cotton reproductive growth, measured as boll number and boll dry weight, was significantly reduced when soil was infested with nematodes. In commercial varieties (Table 2.5), the high level of soil inoculation usually resulted in the least boll number and dry weight. However, in the breeding lines (Table 2.6), there was no significant difference between the mid and the high levels of nematode inoculation relative to cotton reproductive growth.

The differences between the two commercial varieties were often statistically significant, although the difference in dry weight of the shoots was not consistent. Generally, Stoneville 5599BR had significantly higher biomass than FiberMax 960BR in terms of both vegetative and reproductive growth (Table 2.5). There was no difference between PD94042 and 120R1B1 in cotton biomass in trial 2 and reproductive growth in trial 3 (Table 2.6). In trial 3, 120R1B1 had a greater vegetative growth than PD94042.

2.4.5 Photosynthesis

The photosynthetic rate (P_N) was significantly reduced with increased nematode inoculum level in some trials and showed a numeric reduction in the others (Tables 2.7 and 2.8). There was no reduction in photosynthetic rate between commercial varieties in trial 3 with increasing populations of *M. incognita*. An interaction between cotton breeding lines and nematode inoculation levels occurred in trial 2. In this trial, PD94042 demonstrated a significant reduction in photosynthetic rate with increasing populations of *M. incognita*; however, 120R1B1 did not. Other characteristics, to include transpiration rate, stomatal conductance, leaf temperature, and sub-stomatal CO_2 concentration, showed similar trends, although these trends were not always significant. The transpiration rate and stomatal conductance showed trends of reduction with increased populations of *M. incgonita*. An exception to this was observed for the breeding lines in trial 2. Measurements of leaf temperature tended to increase with the exception of breeding lines in trial 2. The trends of sub-stomatal CO_2 concentrations versus population of *M. incognita* were not consistent in different trials, but did significantly increase for commercial varieties in trial 1.

There were no significant differences between FiberMax 960BR and Stoneville 5599BR in any trials for any photosynthesis-related variables (Table 2.7). Breeding line 120R1B1 had a significantly higher transpiration rate, stomatal conductance, and sub-stomatal CO_2 concentration than PD94042 in trial 2 (Table 2.8). However, this difference was not seen in trial 3.

2.5 Discussion

In this study, infestation of soil with eggs of *M. incognita* resulted in significantly higher nematode infection levels in cotton plants, measured as higher root gall ratings and egg counts

compared with the non-infested check. However, the differences in plant measurements between mid and high levels of infestation were usually not significant. This could be an indication that infection by *M. incognita*s on cotton roots was saturated, or nearly saturated, at an infestation level of 6,000 eggs/pot. The egg count data from this study offered further proof that Stoneville 5599BR has some resistance to *M. incognita* because nematode reproduction was suppressed at some level. The resistant breeding line 120R1B1 had significantly higher resistance to *M. incognita* than PD94042 because *M. incognita* reproduction was suppressed 10 to 20 times relative to that on PD94042. The gall ratings and egg counts were not significantly different between the two cotton varieties and between two breeding lines. In some cases, susceptible cultivars FiberMax 960BR and PD94042 resulted in much higher nematode infection than Stoneville 5599BR or 120R1B1 as measured by root gall ratings and egg counts. This confirmed that in susceptible cultivars, southern root-knot nematode can cause more root damage and attain higher levels of reproduction.

It has been suggested that the development of cotton nodes is not influenced by stress before boll set, while plant height is highly influenced by various stresses (Albers, 1993). Therefore the height-to-node ratio is an indicator of the amount of stress that a cotton plant has encountered. Higher height-to-node ratios can indicate that lower stress has occurred in the plants. In all greenhouse trials, *M. incognita* caused a significant amount of stress in cotton plants, reflected as significant reductions in height-to-node ratios. This stress increased significantly as nematode population increased.

In all trials, Stoneville 5599BR was less stressed by *M. incognita* than FiberMax 960BR, because it had significantly better growth than FiberMax 960BR, which proved that Stoneville 5599BR had a better tolerance to *M. incognita* than FiberMax 960BR. Breeding line 120R1B1

appeared to be more stressed by *M. incognita* infection than PD94042 in trial 2 and trial 3 because it usually had a significantly lower height-to-node ratio. However, because 120R1B1 had much less nematode infection than PD94042, the difference in height-to-node ratio is likely due to their genetic difference rather than the influence of *M. incognita*.

The measurement of leaf chlorophyll content provides a measure of photosynthetic capacity related to the nitrogen concentration in the plants (Evans, 1989). As discussed previously, *M. incognita* can stress plants by interfering with water and nutrient transportation (Kirkpatrick et al., 1991; Melakeberhan et al., 1987; Carneiro et al., 2002). Therefore, because chlorophyll content is affected by nitrogen concentration, it was both an indicator of the damage caused by *M. incognita* and an indication of such on photosynthetic activity in the cotton plant. The results from this study showed that infection by *M. incognita* was associated with reduced chlorophyll content in cotton leaves. There was little change in chlorophyll content in plants based upon the mid- and higher infestation levels of *M. incognita*.

Chlorophyll fluorescence is a measure of the excess energy of plants re-emitted as light. It reflects the efficiency of photochemistry and heat dissipation (Maxwell and Johnson, 2000). Therefore the chlorophyll fluorescence signal can be used as an intrinsic probe of photosynthetic function. In this study, chlorophyll fluorescence was only reduced in some trials and the reductions were not always statistically significant. Therefore, the photosynthetic function in cotton may not be affected consistently by *M. incognita*.

Both chlorophyll content and chlorophyll fluoresce responded similarly to levels *M. incognita* inoculation in different varieties and breeding lines. This indicates that *M. incognita* had a similar effect on FiberMax 960BR and Stoneville 5599BR, and on breeding lines PD94042 and 120R1B1. The higher chlorophyll content in FiberMax 960BR may be due to genetic

differences rather than the effect of *M. incognita*, because in previous results, nematode infection reduced chlorophyll content in cotton. Also, Stoneville 5599BR was supposed to have a better tolerance to *M. incognita* than FiberMax 960BR. Chlorophyll content did not differ significantly between the two cotton breeding lines, further confirming that this character was genetic because PD94042 was the recurrent parent of 120R1B1 in a back-cross sequence. These two breeding lines should have similar genetic characters.

Cotton biomass at approximately 65 days after inoculation was significantly reduced when *M. incognita* infected cotton roots in both varieties and both breeding lines. This indicated that *M. incognita* may have interfered with the cotton photosynthetic process, and caused damage in plant growth which led to less biomass. In many cases, root weight was not as significantly reduced as other characters.

Previous literature has shown that *M. incognita* infection can reduce cotton yield (Ortiz et al., 2008). In this study, it was confirmed that *M. incognita* infection reduced cotton boll set and boll weight in all tested varieties and breeding lines. This may lead to reduced cotton yield. Stoneville 5599BR generally had a greater reproductive growth than FiberMax 960BR. These two cultivars usually responded the same to nematode populations. The reproductive growth of breeding line 120R1B1 was less affected by *M. incognita* compared with PD94042, indicating a high resistance in 120R1B1, which may lead to a better yield.

Photosynthetic rate was generally reduced by *M. incognita* infection at around 65 days after inoculation, although the reduction in some trials was not significant. This reduction was often accompanied by an increased leaf temperature, sub-stomatal CO_2 concentration, decreased transpiration rate and stomatal conductance. This symptom was similar to symptoms of drought stress, as drought stress may also lead to increased stomatal closure, leaf temperature and internal

CO_2 content. The drought effect was observed in another study regarding the influence of *M. incognita* on the water relations of cotton in microplots (Kirkpatrick et al., 1995).

In this study, there was no difference between FiberMax 960BR and Stoneville 5599BR in their photosynthetic rates across all nematode inoculation levels. All photosynthetic variables also responded the same in two varieties, indicating the level of resistance did not affect the photosynthetic process of cotton. Breeding line 120R1B1 generally had a higher photosynthetic rate, transpiration rate, stomatal conductance, and lower leaf temperature than breeding line PD94042. These differences were not always statistically significant. But it may indicate that with a higher resistance to *M. incognita*, there was less drought symptom and thus the photosynthetic process was less affected.

Results from this study showed that *M. incognita* infection not only inhibited plant growth, but also damaged plants at a physiological level. Fiber length, which is determined in the elongation stage, requires the deposition of carbohydrate polymers (DeLanghe, 1986). Water pressure inside the developing fiber also has an influence on fiber elongation by regulating the deposition of carbohydrate polymers (Bradow and davidonis, 2000). Therefore, by reducing the production of carbohydrate polymers, which was likely through reducing chlorophyll content and photosynthesis, *M. incognita* could potentially reduce fiber length. To some degree, fiber strength and uniformity can also be influenced by these same stresses (Bradow and davidonis, 2000). Fiber micronaire is determined by a series of carbohydrates produced through photosynthesis that are deposited on the interior walls of the fiber, which increases the value of micronaire (Silvertooth, 1999). Therefore, reduced photosynthesis caused by *M. incognita* infection could lead to reduced fiber micronaire. By reducing the number of cotton bolls, the

competition among bolls for nutrients can be reduced, which could lead to a high micronaire that is not desired.

2.6 Summary and Conclusions

The results of greenhouse trials showed that in response to infesting soil with eggs of *M. incognita*, the height-to-node ratio and chlorophyll content in affected cotton plants approximately 65 days after inoculation was reduced significantly. There was no consistent reduction in chlorophyll fluorescence in plants infected by *M. incognita*. However, a reduced chlorophyll content could lead to the reduced photosynthetic rate that was observed in this study. Photosynthetic rates in three trials at around 65 days after inoculation were negatively affected where infection by *M. incognita* occurred. This reduction was usually accompanied by reduced transpiration rates, reduced stomatal conductance, increased leaf temperatures and sub-stomatal CO_2 concentrations. These effects resembled drought stress in plants.

Cotton biomass as measured in terms of shoot fresh weight and dry weight, root weight and total leaf area were all significantly decreased in plants infected by *M. incognita*. Cotton reproductive growth, measured as number of bolls, and boll fresh and dry weight, was also reduced when the potting soil was infested with *M. incognita* and infection occurred. The differences in growth responses between plants grown in soil with mid-level and high-level nematode populations were usually not significant. This may be an indication that infection and reproduction of *M. incognita* on the cotton roots was saturated or close to saturation at the mid-level nematode population.

There were significant differences between varieties FiberMax 960BR and Stoneville 5599BR regarding their responses to infection by *M. incognita*. Stoneville 5599BR has a higher

level of resistance to *M. incognita* infection than FiberMax 960BR, and this affected cotton growth. Resistance to *M. incognita* in Stoneville 5599BR could also be related to greater suppression of nematode reproductions on roots as compared with susceptible FiberMax 960BR. Breeding line 120R1B1 had a promising resistance to *M. incognita* infection, especially in terms of suppressing nematode reproduction on roots.

Reduced plant growth and physiological characters caused by *M. incognita* could be indicators of suppressed fiber development, because fiber development requires supply of photosynthetic products, and also a normal water pressure and cotton boll numbers for some fiber properties. Resistant cultivars may result in higher fiber quality, because less damage can be caused in these cultivars by *M. incognita*.

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Table 2.1. Effect of different nematode inoculation levels and cotton varieties on gall rating and egg counts.

Trial	Nem level[a]	Gall rating[b]		Egg counts /g root	
		Pooled data[c]		Pooled data	
1	None	0.00 b[d]		0.00 b	
	Medium	4.24 a		2100.02 a	
	high	4.09 a		3004.42 a	
	P value	<.0001		<.0001	
		FM[e]	ST	FM	ST
2	None	0.00 b	0.00 b	0.00 b	0.00
	Medium	4.00 a	2.41 a	8544.71 a	4018.85
	High	4.41 a	2.36 a	14840.77 a	5363.08
	P value	<.0001	<.0001	0.0007	0.1148
		Pooled data		Pooled data	
3	None	0.00 c		0.00 b	
	Medium	2.85 b		5056.02 a	
	High	3.30 a		6330.29 a	
	P value	<.0001		<.0001	
Variety effect		FM	ST	FM	ST
Trial 1	Average	3.03	2.53	2259.68	1202.72
	P value	0.0414		0.0415	
Trial 2	Average	2.80	1.59	7795.16	3127.31
	P value	<.0001		0.0717	
Trial 3	Average	2.50	1.60	4798.53	2792.34
	P value	<.0001		0.0025	

[a] Nematode inoculation level, none = 0 egg/pot, medium = 6,000 eggs/pot, high = 20,000 eggs/pot.
[b] Gall rating was based on a 0-5 index, 0 = no galling, 1 = trace infection with a few small galls, 2 = galling evident on <25% of the roots, 3 = 25-50%, 4 = 50-75% and 5 = >75% of the roots galled.
[c] Data for two varieties was pooled when no significant interaction occurred.
[d] Fisher's Protected LSD(0.05) comparisons among nematode inoculation levels within a trial. Means in a column followed by the same letter are not significantly different.
[e] Cotton variety FM = FiberMax 960BR, ST = Stoneville 5599BR.

Table 2.2. Effect of different nematode inoculation levels and cotton breeding lines on gall rating and egg counts.

Trial	Nem level[a]	Gall rating[b]		Egg counts /g root	
		PD[c]	120	Pooled data	
2	None	0 c[d]	0 b	0.00 b	
	Medium	3.14 b	1.5 a	1650.68 a	
	High	3.91 a	1.73 a	2955.85 a	
	P value	<.0001	<.0001	0.0003	
		PD	120	PD	120
3	None	0.00 b	0.00 b	0.00 b	0.00 b
	Medium	4.25 a	2.20 a	6341.59 a	293.36 a
	High	4.05 a	1.88 a	6878.11 a	272.67 a
	P value	<.0001	<.0001	<.0001	0.1089
Variety effect		PD	120	PD	120
Trial 2	Average	2.35	1.08	2818.65	208.67
	P value	<.0001		<.0001	
Trial 3	Average	2.77	1.36	4406.57	188.68
	P value	<.0001		<.0001	

[a] Nematode inoculation level, none = 0 egg/pot, medium = 6,000 eggs/pot, high = 20,000 eggs/pot.
[b] Gall rating was based on a 0-5 index, 0 = no galling, 1 = trace infection with a few small galls, 2 = galling evident on <25% of the roots, 3 = 25-50%, 4 = 50-75% and 5 = >75% of the roots galled.
[c] Data for each breeding line was shown separately when significant interaction occurred; breeding line PD = PD94042, 120 = 120R1B1.
[d] Fisher's Protected LSD(0.05) comparisons among nematode inoculation levels within a trial. Means in a column followed by the same letter are not significantly different.

Table 2.3. Effect of different nematode inoculation levels and cotton varieties on the areas under the height-to-node ratio, chlorophyll content, and chlorophyll fluorescence progress curve.

Trial	Nem level[a]	AUHNRPC[b] (cm·day)	AUCCPC (day)	AUCFPC (day)
1	None	243.3[c]	1979.2 a[d]	4035.8
	Medium	232.2	1880.5 b	4001.5
	High	223.0	1774.4 c	2995.3
	P value	0.1889	<.0001	0.0531
2	None	156.0 a	1623.7 a	3386.2
	Medium	154.4 a	1530.7 b	3385.7
	High	142.4 b	1484.9 b	3381.7
	P value	0.0048	0.0001	0.9085
3	None	165.4 a	1703.7 a	4009.7 a
	Medium	153.0 b	1570.2 b	3957.2 b
	High	141.1 c	1473.6 c	3975.1 b
	P value	<.0001	<.0001	0.0216

Variety effect		FM[e]	ST	FM	ST	FM	ST
Trial 1	Average	220.3	246.6	1944.76	1803.65	4021.82	3999.89
	P value	0.0003		<.0001		0.1354	
Trial 2	Average	135.1	167.2	1594.2	1498.7	3382.6	3386.4
	P value	<.0001		<.0001		0.6949	
Trial 3	Average	134.3	172.0	1639.0	1526.0	3994.7	3966.6
	P value	<.0001		<.0001		0.0452	

[a] Nematode inoculation level, none = 0 egg/pot, medium = 6,000 eggs/pot, high = 20,000 eggs/pot.
[b] AUHNRPC = area under height-to-node ratio progress curve; AUCCPC = area under chlorophyll content progress curve; AUCFPC = area under chlorophyll fluorescence progress curve.
[c] Data for two varieties was pooled when no significant interaction occurred.
[d] LSD(0.05) comparisons among nematode inoculation levels within a trial. Means in a column followed by the same letter are not significantly different.
[e] Cotton variety FM = FiberMax 960BR, ST = Stoneville 5599BR.

Table 2.4. Effect of different nematode inoculation levels and cotton breeding lines on the areas under the height-to-node ratio, chlorophyll content, and chlorophyll fluorescence progress curve.

Trial	Nem level[a]	AUHNRPC[b] (cm·day)	AUCCPC (day)	AUCFPC (day)
2	None	146.11 a[c]	1615.87 a	3396.77
	Medium	135.29 b	1504.93 b	3396.80
	High	130.68 b	1441.48 b	3397.98
	P value	0.0055	<.0001	0.9886
3	None	157.61 a	1665.85 a	4015.26
	Medium	144.95 b	1540.11 b	3984.85
	High	134.01 c	1489.70 c	4019.37
	P value	0.0002	<.0001	0.0808

Variety effect		PD[d]	120	PD	120	PD	120
Trial 2	Average	143.89	130.33	1513.44	1528.08	3410.51	3383.85
	P value	0.0037		0.6342		0.0053	
Trial 3	Average	147.77	143.28	1550.41	1580.03	4001.34	4011.64
	P value	0.1827		0.0474		0.4970	

[a] Nematode inoculation level, none = 0 egg/pot, medium = 6,000 eggs/pot, high = 20,000 eggs/pot.
[b] AUHNRPC = area under height-to-node ratio progress curve; AUCCPC = area under chlorophyll content progress curve; AUCFPC = area under chlorophyll fluorescence progress curve.
[c] LSD(0.05) comparisons among nematode inoculation levels within a trial. Means in a column followed by the same letter are not significantly different. Data for two breeding lines was pooled when no significant interaction occurred.
[d] Breeding line PD = PD94042, 120 = 120R1B1.

Table 2.5. Effect of different nematode inoculation levels and cotton varieties on plant dry weight, root weight, leaf area, boll number, and boll dry weight.

Trial	Nem level[a]	Dry weight[b]	Root weight	Leaf area	Boll number	Boll dry weight	
		g	g	cm^2		g	
		Pooled data[c]	Pooled data	Pooled data	Pooled data	Pooled data	
1	None	21.53 a[d]	47.68 b	1496.20 a	6.55 a	8.77 a	
	Medium	17.89 b	59.54 a	1485.70 a	4.67 b	7.71 ab	
	High	14.64 c	51.23 b	1320.08 b	3.35 b	2.53 b	
	P value	<.0001	0.0307	0.0201	<.0001	0.0017	
		Pooled data	Pooled data	Pooled data	Pooled data	FM	ST
2	None	15.74 a	29.36 a	1654.03 a	5.77 a	0.50 a	1.41 a
	Medium	11.61 b	26.73 a	1481.85 a	4.00 b	0.17 b	0.70 b
	High	8.72 c	18.41 b	1231.09 b	1.91 c	0.03 b	0.30 c
	P value	<.0001	<.0001	0.0002	<.0001	0.0001	<.0001
		Pooled data	Pooled data	Pooled data	Pooled data	Pooled data	
3	None	17.20 a	50.79 a	1034.66 a	3.70 a	3.19 a	
	Medium	14.19 b	40.22 b	926.64 b	1.85 b	1.62 b	
	High	9.88 c	37.21 b	770.16 c	0.90 c	0.52 c	
	P value	<.0001	<.0001	<.0001	<.0001	<.0001	
Variety effect		FM[e] ST	FM ST	FM ST	FM ST	FM	ST
Trial 1	Average	18.23 17.72	51.43 53.87	1357.02 1501.56	4.16 5.47	4.53	7.88
	P value	<.0001	0.3099	0.0127	0.0006	0.0198	
Trial 2	Average	10.42 13.63	21.02 28.65	1251.55 1659.76	2.39 5.39	0.23	0.80
	P value	0.0008	0.0011	0.0007	<.0001	<.0001	
Trial 3	Average	13.60 13.80	39.68 49.80	855.16 969.54	1.77 2.53	0.55	3.00
	P value	0.2521	0.0086	0.0026	0.0338	<.0001	

[a] Nematode inoculation level, none = 0 egg/pot, medium = 6,000 eggs/pot, high = 20,000 eggs/pot.
[b] Dry weight of the above-ground part of plants.
[c] Data for two varieties was pooled when no significant interaction occurred.
[d] LSD(0.05) comparisons among nematode inoculation levels within a trial. Means in a column followed by the same letter are not significantly different.
[e] Cotton variety FM = FiberMax 960BR, ST = Stoneville 5599BR.

Table 2.6. Effect of different nematode inoculation levels and cotton breeding lines on plant dry weight, root weight, leaf area, boll number, and boll dry weight.

Trial	Nem level[a]	Dry weight[b]		Root weight		Leaf area		Boll number		Boll dry weight	
		g		g		cm^2				g	
		Pooled data[b]		Pooled data		Pooled data		Pooled data		Pooled data	
2	None	15.74 a[c]		34.67		1949.36 a		4.95 a		0.56 a	
	Medium	13.18 b		36.59		1782.31 ab		3.00 b		0.26 b	
	High	11.22 c		30.62		1680.03 b		2.27 b		0.11 b	
	P value	<.0001		0.1693		0.0135		0.0001		<.0001	
		PD[d]	120	PD	120	PD	120	Pooled data		PD	120
3	None	19.6 a	20.9 a	66.6 a	70.1	1218.6 a	1339.1	3.3 a		3.6 a	1.6 a
	Medium	12.7 b	18.4 b	41.7 b	78.6	949.8 b	1315.6	1.2 b		0.8 b	0.5 b
	High	11.3 b	15.60 c	50.2 b	69.9	931.8 b	1216.9	0.7 b		0.0 b	0.1 b
	P value	<.0001	0.0002	<.0001	0.1653	0.0007	0.0750	<.0001		0.0010	0.0257
Variety effect		PD	120	PD	120	PD	120	PD	120	PD	120
Trial 2	Average	13.74	13.02	34.33	33.60	1825.8	1782.0	0.28	0.34	3.24	3.58
	P value	0.3067		0.5025		0.5052		0.5557		0.5005	
Trial 3	Average	14.51	18.39	52.84	72.86	1033.4	1292.3	1.60	1.87	1.47	0.75
	P value	0.001		<.0001		0.0002		0.4575		0.1145	

[a] Nematode inoculation level, none = 0 egg/pot, medium = 6,000 eggs/pot, high = 20,000 eggs/pot.
[b] Dry weight of the above ground part of cotton plants.
[c] Data for two breeding lines was pooled when no significant interaction occurred.
[d] LSD(0.05) comparisons among nematode inoculation levels within a trial. Means in a column followed by the same letter are not significantly different.
[e] Breeding line PD = PD94042, 120 = 120R1B1.

Table 2.7. Effect of different nematode inoculation levels and cotton varieties on photosynthesis.

Trial	Nem[a]	Evap[b]	GS	TL	PN	Ci
		mol $m^{-2}s^{-1}$	mol $m^{-2}s^{-1}$	ºC	µmol m^{-2} s^{-1}	ppm
1	None	3.42[c]	173.41	24.63	14.91 a	212.82 b
	Medium	3.28	166.48	24.74	12.16 b	241.24 a
	High	3.09	147.96	25.57	9.97 b	254.87 a
	P value	0.1819	0.5005	0.1582	0.0047	0.0021
2	None	3.73 a[d]	519.45	21.70	10.20 a	341.32
	Medium	3.22 b	420.32	21.78	8.46 b	340.32
	High	3.30 b	427.41	22.13	8.71 b	336.18
	P value	0.0451	0.1311	0.4888	0.0016	0.3738
3	None	4.56	459.65	25.99	11.47	318.75
	Medium	4.32	417.30	26.10	12.04	310.35
	High	4.21	409.35	26.28	11.57	316.85
	P value	0.4275	0.3464	0.0839	0.8151	0.4869

Variety effect		FM[e]	ST	FM	ST	FM	ST	FM	ST	FM	ST
Trial 1	Average	3.49	3.05	177.28	148.26	25.28	24.72	12.78	11.87	240.47	232.79
	P value	0.9919		0.1989		0.1586		0.4951		0.3240	
Trial 2	Average	3.56	3.27	466.45	445.00	22.15	21.59	9.07	9.18	339.70	338.85
	P value	0.1551		0.6449		0.1463		0.7239		0.8302	
Trial 3	Average	4.52	4.21	445.37	412.17	26.13	26.11	12.35	11.03	314.03	316.60
	P value	0.3012		0.5558		0.8123		0.2021		0.5653	

[a] Nematode inoculation level, none = 0 egg/pot, medium = 6,000 eggs/pot, high = 20,000 eggs/pot.
[b] Evap = transpiration rate; GS = stomatal conductance; TL = leaf temperature; PN = photosynthetic rate; Ci = sub-stomatal CO_2 concentration.
[c] Data for two varieties was pooled when no significant interaction occurred.
[d] LSD(0.05) comparisons among nematode inoculation levels within a trial. Means in a column followed by the same letter are not significantly different.
[e] Cotton variety FM = FiberMax 960BR, ST = Stoneville 5599BR.

Table 2.8. Effect of different nematode inoculation levels and cotton breeding lines on photosynthesis.

Trial	Nem[a]	Evap[b]	GS	TL	PN		Ci	
		mol $m^{-2}s^{-1}$	mol $m^{-2}s^{-1}$	ºC	µmol CO_2 m^{-2} s^{-1}		ppm	
		Pooled data[c]	Pooled data	Pooled data	Pooled data		Pooled data	
2	None	3.76	440.18	22.84	10.30		329.95	
	Medium	3.83	458.32	23.03	10.17		331.64	
	High	3.80	455.05	22.95	9.50		334.23	
	P value	0.9323	0.9004	0.8714	0.0849		0.5129	
		Pooled data	Pooled data	Pooled data	PD[e]	120	PD	120
3	None	3.47 a[d]	1709.9	24.97	15.13 a	16.06	311.5	318.7
	Medium	2.92 b	1992.3	24.72	16.74 a	14.29	301.7	317.3
	High	3.03 b	1142.4	25.28	9.99 b	14.56	326.9	306.5
	P value	0.0286	0.5985	0.5334	0.0040	0.7032	0.1034	0.4904
Variety effect		PD 120	PD 120	PD 120	PD	120	PD	120
Trial 2	Average	3.62 3.97	390.82 511.55	23.14 22.74	9.92	10.06	327.03	336.85
	P value	0.0643	0.0126	0.0758	0.6577		0.0076	
Trial 3	Average	3.20 3.08	1510.3 1719.4	25.26 24.71	13.95	14.97	313.37	314.17
	P value	0.4696	0.6732	0.0586	0.4505		0.8791	

[a] Nematode inoculation level, none = 0 egg/pot, medium = 6,000 eggs/pot, high = 20,000 eggs/pot.
[b] Evap = transpiration rate; GS = stomatal conductance; TL = leaf temperature; PN = photosynthetic rate; Ci = sub-stomatal CO_2 concentration.
[c] Data for two breeding lines was pooled when no significant interaction occurred.
[d] LSD(0.05) comparisons among nematode inoculation levels within a trial. Means in a column followed by the same letter are not significantly different.
[e] Breeding line PD = PD94042, 120 = 120R1B1.

CHAPTER 3

THE EFFECT OF SOUTHERN ROOT KNOT NEMATODE (*MELOIDOGYNE INCOGNITA*) ON COTTON GROWTH, YIELD, AND FIBER QUALITY IN THREE FIELDS WITH SITE-SPECIFIC MANAGEMENT[1]

[1] Lu, P., Kemerait, R. C., Jr., Davis, R. F., and Perry, C. To be submitted to *Journal of Nematology*.

3.1 Abstract:

Southern root knot nematode [*Meloidogine incognita* (Kofoid & White) Chitwood] is a major parasite on cotton (*Gossypium hirsutum L.*), causing considerable yield loss. At three field sites, different types and rates of nematicides were applied in an effort to create different populations of *M. incognita* in 2008 and 2009. Each field was delineated into two site-specific management zones. In all three fields in both years, Telone II significantly reduced soil populations of *M. incognita*, root gall ratings, and nematode reproduction on the roots and significantly improved plant growth and yield as compared with Temik 15G and seed treatments. Differences between variable rates of the same nematicides or different nematicide seed treatments were not statistically significant. Most measurements of fiber quality were not significantly or consistently affected by treatments across trials and years. The effects of management zones were usually not significant but differed from field to field.

Keywords: Fiber quality, Management zones, *Meloidogyne incognita*, Southern root-knot nematode, Precision agriculture, Yield

3.2 Introduction

Cotton (*Gossypium hirsutum L.*) is an oilseed and fiber crop. It is grown in more than seventy countries and is the single most important fiber crop worldwide. No crop competes with it in the potential of value-added processing (Basra, 1999). Cotton is also one of the oldest cultivated crops. It has been associated with human activity since before recorded history.

The U.S. is the third-largest producer of cotton in the world. Cotton production in the U.S. has increased 66% in the past 40 years (Mitchell, 2009). In recent years, the U.S. has produced about 20 percent of the world's annual supply (Mitchell, 2009).

Root knot nematodes occur worldwide and attack a diversity of crops. They cause considerable yield losses and affect the quality of the product, either by directly damaging plants (Kirkpatrick and Sasser, 1984) or by predisposing them to infection by fungal and bacterial pathogens (Powell, 1971).

The southern root knot nematode [*Meloidogyne incognita* (Kofoid & White) Chitwood] is found in all cotton production regions in the U.S. and is the most widely distributed nematode parasite of economic importance of the crop (Thomas and Kirkpatrick, 2001). It is considered the major yield-limiting plant-parasitic nematode across the U.S. Cotton Belt; approximately twice as much yield loss is attributed to *M. incognita* as to all other nematode parasites of the crop (Koenning et al., 2004). The estimated yield loss of cotton caused by *M. incognita* in the U.S. was 2.4% in 2007, which was greater than for any other cotton disease. This damage resulted in a loss of more than 106,000,000 kg of lint (Cotton Disease Loss Committee, 2008). In Georgia in 2007, *M. incognita* caused an estimated 6% reduction in yield resulting in a loss of 25,000,000 kg of lint (Cotton Disease Loss Committee, 2008).

Damage caused by plant-parasitic nematodes is distinct from other plant diseases because there are no unique symptoms on the above-ground portions of the plants. Nematodes are often unevenly distributed in the soil; therefore the symptoms associated with damage from nematodes may occur in irregular patches in the field (Beltwide Cotton Committee, 2003). These patches can be either small and limited in number, or large and widely distributed. Damaged plants may exhibit symptoms ranging from mild to severe stunting depending on the level of infestation and a reduced rate of development. Foliage may also show symptoms of nutritional deficiency (Kirkpatrick et al. 1995). In the most severe cases, plants may die before maturation. The nematodes also interact with the Fusarium wilt pathogen (*Fusarium oxysporum* f.sp. *vasinfectum*), which leads to wilting and brown discoloration or necrosis of the vascular tissue of the lower stem (Beltwide Cotton Committee, 2003).

Below-ground symptoms caused by *M. incognita* on cotton can be much more diagnostic than above-ground symptoms. Visible galls or "knots" often appear on cotton roots (Bridge and Page, 1980). Swellings of the infected root tissue can be found on the cotton tap root and the lateral roots: however, the galls on cotton may not be as easy to observe as those on vegetables such as tomatoes. Galls are easier to detect if cotton plants are carefully dug (not pulled) from the soil. Also, the fine lateral roots need to be handled carefully when rinsed with water to remove soil (Beltwide Cotton Committee, 2003).

The tap root and its lateral roots are of vital importance to the cotton plant. A few galls on these roots can disrupt the normal flow of water and nutrients to the leaves and developing bolls, which can significantly reduce the yield of cotton (Bird and Loveys, 1975; Kirkpatrick et al., 1991; McClure, 1977).

Previous research has focused on the management of *M. incognita* to improve cotton yield. Very little data has documented the impact of *M. incognita* on cotton fiber quality. However, cotton fiber quality is a very important issue to the growers and to the textile industry. Fiber quality is a set of measurements that describe a sample of fibers extracted from a bale of cotton (Bradow and Davidonis, 2000). These measurements include length, uniformity, strength, micronaire, color grade, trash, leaf grade, preparation, and extraneous matter (USDA, 2001). They are compared with a set of standards from the U.S. Department of Agriculture (USDA) and are used to determine price premiums and discounts.

In recent years, cotton lint quality has become increasingly important because of the requirements from textile mills for fiber quality, the use of high-volume instrument (HVI) testing, the occurrence of discounts due to unfavorable fiber characteristics, and depressed cotton markets (Silvertooth, 1999). Georgia is the third-largest cotton producer in the U.S. In the 1990s and early 21st century, Georgia produced cotton had the fiber quality second only to that produced in California, but now certain fiber quality parameters have not only fallen below levels found in western U.S cotton, but also below the average fiber quality of other southern and southeastern states (Bradow and Davidonis, 2000).

There are four general phases in cotton fiber development: 1) initiation, 2) elongation, 3) thickening, and 4) desiccation (maturation). Fiber length is determined in the elongation stage, which occurs about 21 days after flowering. During this stage, a thin cell wall of carbohydrate polymers is deposited allowing the fiber to elongate (DeLanghe, 1986). Water pressure inside the developing fiber has an influence on fiber elongation through regulating the deposition of carbohydrate polymers (Bradow and Davidonis, 2000). Therefore, if stresses occur during this stage (which are mainly associated with water stress or potassium deficiency), fiber length can

be reduced (Silvertooth, 1999). To some degree, fiber strength and uniformity can also be influenced by these same stresses (Bradow and Davidonis, 2000).

The thickening process of cotton fibers may overlap with elongation to some extent. During this stage, a series of carbohydrates produced through photosynthesis are deposited on the interior walls of the fiber, which increases the value of micronaire (Silvertooth, 1999). If this development is stopped prematurely, the micronaire value will decrease and finer fibers will occur. New bolls that are set on the plant usually have some of the greatest demand for carbohydrates. These bolls draw carbohydrates from the older bolls, which prevents the development of high-micronaire fibers on older bolls (Silvertooth, 1999).

The maturation of fiber occurs after the boll has opened and the metabolically inactive fibers dry. There is no quality measurement directly related to the maturation process. The fiber quality within a boll is at its utmost on the day of boll opening (Bradow and Davidonis, 2000). Therefore harvesting should be as close to physiological maturity as possible to enhance the quality of the crop produced.

Environmental factors and management factors can significantly alter cotton fiber quality. For example, reduced light (cloudy) conditions result in the production of weaker fibers with reduced micronaire (Pettigrew, 2001). Early defoliation can also reduce micronaire in cotton (Snipes and Baskin, 1994).

Biological factors such as plant-pathogenic fungi and pests have also been studied. Cotton root rot, caused by the soilborne fungus *Phymatotrichum omnivorum*, can reduce fiber length and fineness significantly (Mulrean, 1984). It has also been well documented that by feeding on cotton bolls, stink bugs can decrease fiber quality as indicated by a reduction in almost all HVI-measured variables (Roberts et al., 2005). *Meloidogyne incognita*, a root-feeding

plant parasite, is another factor that could potentially affect fiber quality. *M. incognita* draws nutrients and water from cotton plants and exacerbates moisture, nutrient, and other kind of stresses and also fungal infection, thus having the potential to indirectly cause damage to fiber development.

In previous studies, several fiber properties were documented to be affected by moisture and nutrient stress, reduced or delayed cotton growth, which in other studies, were associated with infection by *M. incognita*.

Fiber length and staple can be influenced by several factors including variety, temperature, water stress, nutrient deficiencies, and ginning practices (Bradow and Davidonis, 2000). Water relationships and irrigation practices have been studied primarily in relation to yield. One study conducted in the early 1980s indicated that fiber length was not impacted until water stress was such that yields were limited to less than 706.1 kg/ha (Grimes and Yamada, 1982). The interaction between *M. incognita* and water stress in cotton has also been studied. The results indicated that in susceptible cultivars, infection by *M. incognita* may decrease the movement of water from roots to leaves. The decrease in root flux caused by nematodes is equal to that induced by severe water deficit stress (Kirkpatrick et al., 1991). Other studies have demonstrated that water stress early in the bloom period had a less negative impact on fiber length than water stress late in the bloom period (Hearn, 1976; Marani and Amirav, 1971; Shimishi and Masani, 1971;). Sensitivity of fiber elongation to severe water stress is apparently due to the physiological and mechanical processes of cell expansion (Hearn, 1994). As *M. incognita* infection gets more and more severe later in the season, the resulting water stress in the cotton plant may lead to a reduction in fiber length.

The possible impact of *M. incognita* on fiber strength is not as direct as the impact on fiber length. Firstly, fiber strength has a positive relationship with canopy sunlight absorption. There is evidence that *M. incognita* can reduce plant leaf area, e.g., in tomato (Loveys and Bird, 1973). Therefore by reducing cotton leaf area, *M. incognita* may affect fiber strength. Secondly, studies have indicated that heat accumulation during the flowering period can also affect fiber strength. Fiber strength was greatest in bolls that developed from flowers produced during the first 4 to 6 weeks of flowering. Flowers that opened during the latter 2 weeks of the flowering period produced bolls with the lowest fiber strength (Jones and Wells, 1997). Therefore, *M. incognita* may reduce fiber strength by delaying cotton development and flowering.

Authors in previous study have suggested several factors that could influence fiber micronaire. Significant differences in micronaire among commercially available varieties have been examined. A fiber thickens as cellulose is deposited inside the fiber cell. If photosynthesis of cotton is affected by *M. incognita*, as confirmed in tomato plants (Loveys and Bird, 1973), the lack of carbohydrate production may lead to a lower micronaire value. However, micronaire values that are too high can also result in loss of profit. It is important for cotton to produce sufficient bolls later in the season to either compete for carbohydrates or to produce sufficiently lower micronaire fiber. This fiber can be blended with the higher micronaire fiber to reduce overall readings (Silvertooth, 1999). Therefore, the reduced growth and boll setting in cotton caused by *M. incognita* (Kirkpatrick et al., 1995; Walker et al., 1998) may affect the blended micronaire value.

As discussed previously, water stress can be caused by *M. incognita* (Kirkpatrick et al., 1991). In other studies, it has also been shown that water deficiency in cotton can affect some properties of cotton fiber. Moisture stress later in the season was found to reduce fiber length and

micronaire (Marani and Amirav, 1971). The reduction in fiber length due to water stress was also observed in a later study. Although other characters were not consistently affected, the distribution of bolls was consistently and significantly affected by irrigation (Pettigrew, 2004).

Studies on the effect of *M. incognita* on nutrient concentration in plants show that a change in concentration of the nutrients in the plant is likely one of the first effects of the nematode on host physiology. These changes in nutrient concentration alter host metabolism and contribute directly or indirectly to the chlorosis and premature leaf abscission in soybean (Melakeberhan et al., 1987). This is also true according to another study in soybean. The uptake of nitrogen, phosphorus, and calcium was affected by *M. incognita* (Carneiro et al., 2002). Many nutrients have also been reported to have effects on fiber quality. For example, leaf nitrogen during the boll maturation period had significant positive correlations with fiber length and negative correlations with micronaire (Reddy et al., 2004). Potassium is also a very important element for cotton and has positive correlations with fiber length, micronaire index, fiber strength, and fiber length uniformity ratio (Cassman et al., 1990).

Results of greenhouse studies from Chapter 2 showed that *M. incognita* infection significantly reduces cotton plant growth, and some physiological characters including chlorophyll content and photosynthesis. This reduction during plant growth could lead to interrupted fiber development, because it requires sufficient photosynthetic products and normal water pressure.

Approaches to effective nematode control include crop rotation, field sanitation, cover crops, varietal resistance and nematicides. Nematode population densities can be reduced by selecting rotation crops that are not hosts to *M. incognita*, or by planting resistant cultivars (Rich and Kinloch, 2005). To date, no *M. incognita*-resistant cotton cultivar is commercially available.

Several cotton cultivars have been reported to have some level of tolerance to the Fusarium wilt/*M. incognita* disease complex. However, these cultivars do not show significant resistance to the nematodes, although some are resistant to Fusarium wilt (Beltwide Cotton Committee, 2003). Biological approaches for nematode control have been studied but have not been thoroughly explored in cotton production systems (Starr et al., 2007).

Chemical nematicides are widely used to control *M. incognita*. Three nematicide management strategies are currently widely used in the U.S. (Koenning et al., 2004), including the application of aldicarb at rate of 0.8-1.2 kg/ha in the planting furrow; preplant soil fumigation using either 1,3-dichloropropene or metam-sodium (Starr et al., 2007); and the supplemental use of either aldicarb applied as a side-dress during the first third of the season or a foliar application of the carbamate oxamyl (Lawrence and Mclean, 2000, 2002). Out of all these strategies, at-planting application of aldicarb is perhaps the most universal nematicide strategy in the U.S. Aldicarb is applied on 20 to 30% of the cotton hectarage each year (Koenning, et al., 2004). In severely infested fields, nematicides can reduce the nematode population by more than 50% (Beltwide Cotton Committee, 2003). Using chemical nematicides can provide cotton with a zone of protected soil in which roots can develop for 4 to 6 weeks with reduced damage from nematodes. By protecting the crop during early development, yield losses can be reduced substantially even though nematodes may penetrate the roots during the latter part of the season (Beltwide Cotton Committee, 2003). Seed treatments have shown some promising results in protecting emerging roots from nematode infection (Monfort et al., 2006), but may not be sufficient in fields with high nematode population densities.

In Georgia, commonly used practices for cotton growers are the application of the soil fumigant Telone II (1,3-dichloropropene) before planting and application of granular

nematicides such as Temik 15G (aldicarb). However, the cost of nematicides may cause growers to use them sparingly in their production system (Ortiz et al., 2008).

Like other soil-borne plant pathogens, *M. incognita* is present in the soil in an aggregated pattern and the damage it causes often appears as patches in the field. This aggregated distribution is affected primarily by the variability of soil properties. It has been shown that out of 26 edaphic components including soil texture, acidity, pH, and others, 50% of the variability in nematode density was related to high levels of clay, organic matter, low copper concentration, and small changes in percent soil moisture (Noe and Barker, 1985).

To offer a more cost-effective tactic for use of nematicides on cotton, a management zone strategy targeting areas with damaging nematode populations for treatment with nematicides has been studied. The management zone strategy is based on the fact that *M. incognita* is present in the soil in aggregated patterns due to the variability of soil texture in a field, as *M. incognita* prefers coarse-textured, sandy soils. By using site-specific management (SSM), high-rate nematicides will only be aimed at zones that potentially have higher nematode populations. A lower rate or less expensive nematicide will be applied in low nematode risk zones. Hopefully, the efficacy of control of nematodes can be improved and expenses can be reduced.

In 2001, a strong correlation between Columbia lance nematode densities and sand content was observed from electrical conductivity data that was used to map within-field spatial distribution of soil texture (Khalilian et al., 2001). This indicated promise to application of SSM to Columbia lance nematodes. In recent years, SSM has also been studied for *M. incognita* in cotton fields. The data indicated that edaphic features can be used as indirect indicators of high-risk, nematode-prone areas (Ortiz et al, 2006). In 2007, it was found that different nematicide rates had significantly different effects on cotton yields within different risk zones (Ortiz et al.,

2008). These findings support the strategy of variable-rate nematicide applications based on root knot nematode risk management zones. The goal of using SSM in this study was to determine whether cotton fiber quality may also respond differently to nematicide application in different management zones.

The objective of this chapter was to (1) examine the effect of *M. incognita* infection on cotton growth, lint yield, and fiber quality under field condition; and (2) determine whether these effects are influenced by different risk management zones.

3.3 Materials and Method

The experiments were conducted at three commercial fields 2008 and again in 2009. The three field sites included a field at the Perryman farm in Colquitt County and two fields ("east" and "west") at the Windhausen Farm in Mitchell County. These three fields had been in cotton for a long time, were non-irrigated, and had histories of significant yield losses to *M. incognita*. The tillage method used at the Perryman farm was conventional where the land was ripped and bedded prior to planting. Conservation/strip tillage was practiced on the Windhausen farm where the fields were planted to a rye cover crop during the winter months. Delta and Pineland 555BR was the only cotton variety planted in these three fields over the course of this study. This variety has no known resistance to the southern root-knot nematodes. The planting date for Perryman farm was 11 May 2008 and 14 May 2009. For Windhausen farms, the planting date was 24 May 2008 and 13 May 2009.

Six treatments were applied at Perryman farm in order to assess the efficacy of the commercially available nematicides and to attempt to create different nematode population levels in the soil with which to challenge the cotton plants. The treatments included : (T1) Temik 15G

(aldicarb 3.36 kg/ha); (T2) Temik 15G (aldicarb 6.72 kg/ha); (T3) Telone II (1,3-dichloropropene 28.1 L/ha) + Temik 15G (aldicarb 3.36 kg/ha); (T4) Telone II (1,3-dichloropropene 56.12 L/ha) + Temik 15G (aldicarb 3.36 kg/ha); (T5) AVICTA Complete Cotton and (T6) AERIS Seed-Applied System + Trilex. Temik 15G at 3.36 kg/ha is the standard rate of this product used for early-season management of thrips. At this rate, Temik 15G is thought to have only minimal nematicidal activity. Fumigation with Telone II offers the best protection for the developing cotton plant from plant-parasitic nematodes but requires additional equipment and cost. The addition of 3.36 kg/ha Temik 15G to the two Telone treatments was for thrips control. Seed treatment AVICTA Complete Cotton is a combination of three separately registered products, including AVICTA (abamectin), Cruiser (thiomethoxam), and Dynasty CST (azoxystrobin, fludioxonil and mefenoxam). AERIS Seed-Applied System + Trilex is a combination of the insecticide imidacloprid at 0.375 mg a.i./seed and nematicide thiodicarb at 0.375 mg a.i./seed with an optional fungicide package of Trilex Advanced (trifloxystrobin, triadimenol, and metalaxyl). AVICTA Complete Cotton and AERIS Seed-Applied System were not included in trials at the Windhausen farm. Therefore, there were only four treatments included in these two fields.

At each site, treatments were applied in strips across the entire length of the field. At the Perryman farm, the treatment strips were four rows wide. On the Windhausen farm treatment strips were eight rows wide. Each of the three fields was delineated into two management zones based upon fuzzy clustering of various surrogate data for soil texture. The high-risk zones in each field tended to have sandier soil which is preferred by *M. incognita*, and are therefore expected to have higher *M. incognita* populations. Soil in the low-risk zones has a lower percentage of sand and is believed to have lower nematode populations. The methodology for

management zone delineation was developed using data collected in 2005 and 2006 from 11 cotton fields (Ortiz et al., 2007). The surrogate data for soil texture used in management zone delineation included terrain elevation and slope, normalized difference vegetation index (NDVI) calculated from bare-soil spectral reflectance, and apparent soil electrical conductivity (ECa). These data was collected in the previous year after harvest. EC values were measured using VERIS 3100 implement (Veris Technologies, Salina, KS), and elevation data was collected with a Trimble AgGPS 214 real-time kinematic GPS receiver (Trimble, Sunnyvale, CA) mounted on the tractor pulling the VERIS 3100 implement.

Within each field an experiment was established with a completely randomized design in 2008 and a randomized complete block design in 2009. In 2008, treatments strips and subplots were randomly arranged within the entire field. In 2009 the treatment strips and subplots were randomly assigned within complete blocks. For each field, there were six treatment blocks across the field. In 2008, five sampling plots were randomly assigned for each treatment within each management zone in the area where a certain treatment was applied (Figure 3.1). In 2009, the sampling plots were assigned in blocks, where all the treatments for one replication were gathered in one block. There were three replicated blocks in each management zone in 2009 (Figure 3.2). The width of each sampling plot was four rows at Perryman farm and eight rows at the Windhausen farm. The length of sampling plots was 15.2 m in 2008 and 30.5 m in 2009.

Populations of southern root-knot nematodes were determined by collecting soil samples from the center two rows of each subplot using soil probes. The soil samples were then sent to the University of Georgia's Nematology Lab in Athens for processing. Nematodes were extracted from 100 cm^3 soil using centrifugal-flotation method (Jenkins, 1964) and *Meloidogyne*

juveniles were counted using a dissecting microscope. Soil samples were taken three to four times in each field per season on a monthly basis (Table 3.1)..

Cotton shoot heights were measured approximately every 2 weeks during the first half of growing season and until the cotton plants reached the "cut-out" developmental stage. For each sampling plot, five plants were randomly selected and measured. At approximately 4 and 8 weeks after planting, plant samples were taken from each field for further measurements. Five random plants from the center two rows of the sampling plots were dug carefully from the soil, separated into shoots and roots, and then stored in plastic sampling bags.

The measurements taken at first destructive sampling included:

(1) Height-to-node ratio: This was calculated by dividing shoot height by the number of nodes. The number of nodes was recorded by counting the number of leaves that are bigger than a U.S. quarter coin above the cotyledons on the main stem;

(2) Shoot dry weight: The five plant shoots from each sampling plot were packed into one paper bag and dried in oven at 60˚C for 2 days before dry weight was measured;

(3) Root gall rating: This was based on a 0-5 index, 0 = no galling, 1 = trace infection with a few small galls, 2 = galling evident on <25% of the roots, 3 = 25-50%, 4 = 50-75% and 5 = >75% of the roots galled (Kinloch, 1990).

(4) Egg counts: *Meloidogyne* eggs were extracted with 0.625% NaOCl from cotton roots by shaking for 3 min (Hussey and Barker, 1973) and were counted microscopically.

For the second destructive sampling, only cotton roots (n = 5) were sampled because the tops were too large to handle. Therefore, root weight, gall rating, and egg counts were measured at the second destructive sampling.

Data for variables that were measured repeatedly were transformed into "areas under the variable progress curves" using the trapezoidal method. The formula used to calculate the areas was the same as the formula for the area under disease progress curves, which is $\sum_{i=1}^{n-1}\left(\frac{y_i + y_{i+1}}{2}\right)\left(t_{i+1} - t_i\right)$, where t is the time (in days) after planting at each measurement, y is the value of the variable that was being measured, and n is the number of measurements over time. The areas under the progress curves from each treatment were than analyzed using ANOVA and LSD tests using the PROC GLM procedure in SAS (SAS Institute, Cary, NC). The data for Windhausen East and West farms were combined for analysis. The ANOVA and LSD tests for the combined data were conducted using PROC MIXED procedure in SAS.

During the 2009 growing season, cotton leaves from the fifth node on the main stem were collected and sent for nutrient concentration analysis. Soil was collected mid-season for nutrient analysis on 4 September 2008 for Perryman farm and on 11 September 2008 for Windhausen West and East farms. In 2009, soil was collected for nutrient analysis on 15 September for all three fields.

Toward the end of the season, the location of all sampling plots at each of the three fields was mapped using GPS in order to monitor cotton yields accurately at harvest from sampling plots. At harvest the spatial variability of cotton lint yield was recorded using an Ag Leader cotton yield monitor system (AgLeader Technology, Ames, IA) installed on a Model 9965 four-row John Deere picker. Within each subplot, samples were caught from the picker using a fishing net and later processed with a table gin. For larger plots, samples were taken from the cotton picker and ginned at the University of Georgia microgin in Tifton. Cotton lint was then sent to the USDA cotton classifying office in Macon, GA, for quality testing.

To evaluate the economic impact of *M. incognita* on cotton production with respect to fiber quality, the lint price per pound was calculated using the Southeast base price from the spot cash market (averaged December 2008 and 2009 data) plus the premiums and/or minus the discounts for quality. The average base price equaled $0.6549/lb. In 2008 the price included a loan deficiency payment because it was below the loan rate of $0.52/lb. In addition to lint price, the adjusted revenue was also evaluated. The adjusted revenue took many important economical influencers into account, including nematicide application, variety, tillage, labor, yield, fiber quality, etc. Both lint price and adjusted revenue were analyzed with ANOVA and LSD tests using PROC GLM in SAS.

The effects of treatments were examined. Principal component analysis was also conducted in SAS using PROC PRINCOMP to analyze the relation between response variables (yield and fiber quality variables) and explanatory variables (plant growth, nematode population and reproduction, soil nutrient contents, etc.). The biological interpretation of each principal component was determined by finding the explanatory variables that had the highest absolute values of eigenvectors, usually above 0.25 to 0.30. Regression analysis of the most important principal components, which together explain over 70% of the total variation, was conducted against the response variables of interest.

3.4 Results

3.4.1 *Meloidogyne. incognita* populations, disease severity, and reproduction

Meloidogyne incognita populations at the Perryman farm over 2 years showed similar differences among treatments and between zones (Table 3.2). Where either rate of Telone treatment was applied, *M. incognita* juvenile counts were significantly lower than in other

treatments. However in 2009, the lower rate of Telone was not statistically different from the higher rate of Temik 15G. There was generally no significant difference among seed treatments and Temik treatments with regard to soil nematode populations. Typically the differences between soil nematode populations in the high and low rates of Telone were not significant in either year. Differences between plots planted to seed treatments and plots planted to Temik treatments were more obvious in terms of root gall ratings and egg counts per gram of roots, where use of seed treatments resulted in greater gall ratings and egg counts than in other treatments. Roots from plots fumigated with Telone II had the lowest gall ratings and egg counts. The differences of soil nematode populations, root gall ratings and egg counts were mainly among nematicide types rather than between different rates of the same nematicide. Differences between Temik and seed treatments were not as obvious, especially with the lower rate of Temik; most of the time it was not statistically separated from seed treatments.

At the Windhausen farm (Table 3.3), soil *M. incognita* populations were also significantly lower with Telone treatments and higher with Temik treatments. This difference was numerical but not statistically significant in 2008. The gall ratings and egg counts were also significantly greater with Temik treatments than Telone treatments. The difference between different rates of nematicides was not significant for any variable in either year, which was consistent with the results from the Perryman farm.

3.4.2 Cotton growth

The plant vegetative growth measured as higher shoot height, height-to-node ratio, and shoot dry weight at Perryman farm in both 2008 and 2009 (Table 3.4) was consistently the best with Telone treatments. Although root weight was numerically higher with Telone treatments, the difference was not statistically significant in either year. The differences between plants

from plots treated with Temik and plots with seed treatments were not significant in shoot height measurements. However, the height-to-node ratios and dry weights were significantly higher with Temik treatments than with seed treatments. There was no difference in plant growth measurements between rates of a specific nematicide.

At the Windhausen farm fewer differences were found in cotton growth among plants measured from plots treated with nematicides (Table 3.5). Shoot height measured in 2009 was the only character that differed significantly. Plant heights from plots fumigated with Telone II were significantly higher than were plants from plots receiving Temik treatments.

3.4.3 Cotton yield

At the Perryman farm, cotton lint yield was significantly greater where the higher rate of Telone II was applied than where other treatments were applied in 2009 (Table 3.6). Yield collected from plots fumigated with the lower rate of Telone II was not statistically different from yields of Temik and seed treatment plots. There was no difference between Temik and seed treatments in lint yield. In both years at Perryman farm, fumigation with the higher rate of Telone resulted in the highest lint yield; use of seed treatments resulted in the lowest yield across all sampling plots, although in 2008 this was not statistically significant. Across the entire field, Telone treatments resulted in significantly greater yield in both years (Table 3.8). The difference between Temik and seed treatments was not significant in either year.

At the Windhausen farm, the results of lint yield from 2008 and 2009 were seemingly opposite to each other (Table 3.7). In 2008, the lint yield was the greatest in plots fumigated with the higher rate Telone II. The two Telone treatments also resulted in the highest yield across the entire fields (Table 3.9). However in 2009, the high rate of Telone resulted in a significantly

lower lint yield than any other treatments. Across the entire fields, cotton lint yield was not significantly affected by nematicide treatments.

3.4.4 Fiber quality

In both 2008 and 2009 there was no apparent interaction between fiber quality and risk management zone at the Perryman farm (Table 3.6). Therefore, for most of the fiber properties, data from the two management zones was pooled together for analysis. The only fiber properties that potentially had interactions between nematicides and management zones were trash content in 2008 and fiber yellowness in 2009. Trash content in 2008 was significantly higher in plots treated with AVICTA Complete Cotton in the low-risk zone. Trash content was lowest in plots treated with the high rate of Temik. Fiber yellowness in 2009 was the lowest in lint from plots with AVICTA Complete Cotton. None of these trends at Perryman farm were consistent.

Fiber quality data from fields at the Windhausen farm was pooled together for analysis as there appeared to be no interaction between fields and treatments or management zones. In the 2008 trials (Table 3.8), none of the fiber properties appeared to have interactions between treatments and management zones. Fiber reflectance and trash content differed significantly with different nematicide treatments. Use of the low rate of Temik 15G resulted in the highest fiber reflectance and use of the high rate of Telone resulted in the highest trash content. In the 2009 trials, color grade, micronaire, strength, and fiber yellowness all seemed respond differently to nematicide treatments in different management zones. Micronaire was higher with Telone treatments only in the high-risk zone, and fiber yellowness was higher with Temik treatments only in the high-risk zone. Fiber strength was lower with the low rate of Telone than any other treatments in the low-risk zone.

Cotton fiber quality from across the entire Perryman farm did not differ significantly among nematicide treatments in terms of any fiber properties in either year (Table 3.8). At the Windhausen farm (Table 3.9), fiber yellowness was significantly higher where the high rate of Telone was applied and was significantly lower with the high rate of Temik in 2008. In 2009, fiber reflectance was significantly lower with the low rate of Telone. Other fiber properties were all not significantly different with different nematicide treatments.

3.4.5 Principal component analysis

The eigenvectors for the combined 2008 trials from the Perryman field and the Windhausen West and East fields (Table 3.10) showed that plant growth variables (shoot height, dry weight, number of nodes, etc.) contributed the most to principal component 1. Therefore, principal component 1 was considered a plant growth component. Four nematode variables and two soil variables contributed the most to principal component 2; therefore principal component 2 was mainly a nematode component. Principal component 2 also included some soil and nematode combined effects. Principal component 3 was mainly a soil component. Plant growth variables and nematode and soil variables had a combined effect on principal component 4. Principal component 5 was again mainly a nematode component. These five principal components together explained 71.7% of the total variation.

According to the results of regression analysis of yield and fiber quality against the first five principal components (Table 3.11), plant growth had significant positive effect on cotton yield and several cotton fiber properties to include staple, strength, length and uniformity, but had a negative effect on micronaire. Principal component 2 had significant positive effect on yield, fiber staple, and length, but had a negative effect on micronaire. The other nematode

component, principal component 5, had a negative effect on yield, fiber staple and length, and many other fiber properties, although the effects were all not significant. Principal component 3 and 4 were both plant growth and soil combined components and they had significant effects on yield and many of the fiber properties. However, the effect between component 3 and 4 were opposed to each other.

The principal component analysis for trials conducted in 2009 (Table 3.12) showed that plant growth variables contributed significantly to principal component 1. Therefore, principal component 1 was considered a plant growth component. Principal component 2 was a plant growth and leaf nutrient combined component. Principal component 3 was a soil component. Contributions to principal component 4 came mainly from plant growth and leaf nutrient variables, but also one nematode variable. Principal component 5 was again a leaf nutrient component. Principal component 6 was a nematode component and principal component 7 was a nematode plus leaf and soil nutrient component. These seven components together explained 70.3% of the total variation.

Principal components 1, 2 and 3 had significant effect on yield (Table 3.13). In 2009 the plant growth component and soil component had a significant negative effect on cotton lint yield. Many of the fiber properties were positively affected by the first or second component. These were plant growth and plant growth plus leaf nutrient components. The nematode component had no significant effect on either cotton yield or fiber quality in 2009.

3.4.6 Economic impact

The results of lint price at Perryman as well as Windhausen West and East fields in 2008 and 2009 (Table 3.14) showed that in all three fields, in both management zones, and in both

years, the price of lint per pound did not differ with different treatments. The adjusted revenue for Perryman, Windhausen West and East fields in 2008 and 2009 (Table 3.15) showed that at Windhausen West and East fields, the adjusted revenue was usually significantly higher with Temik treatments than Telone treatments in 2009. However in 2008, although only numerically, the low rate of Temik usually resulted in the highest adjusted revenue, but most of the differences were not significant. At Perryman farm in both years, no significant change in adjusted revenue was found due to different nematicide treatments.

3.5 Discussion

At the Perryman, Windhausen West and Windhausen East fields, different levels of *M. incognita* populations were successfully created. The effects of nematicides were significantly different in terms of area under nematode counts, gall rating, and egg count progress curves. However, different rates of the same nematicide or different seed treatments were not significantly different. Generally speaking, areas under nematode counts, cotton root gall rating, and egg counts progress curves in both years were the lowest from plots fumigated with Telone II and were the highest where seed treatments were used. Therefore, a higher *M. incognita* population in soil resulted in higher levels of infection and nematode reproduction on cotton roots. Telone treatments significantly suppressed nematode infection and reproduction, and this result was achieved with even the low rate of Telone. The high rate of Telone did not significantly increase this suppression.

It has been suggested that the development of cotton nodes is not influenced by stress before boll set, whereas plant height is highly influenced by various stresses (Albers, 1993). Therefore the height-to-node ratio is an indicator of the amount of stress that a cotton plant has

encountered. Higher height-to-node ratios can indicate that lower stress has occurred in the plants. The results from measurement of height-to-node ratios showed that Telone treatments significantly reduced the stress in cotton plants that was caused by *M. incognita* as compared with Temik 15G and seed treatments. This appears to be true as use of Telone II resulted in the highest height-to-node ratios.

With a better *M. incognita* control throughout the growing season, the higher rate of Telone II significantly increased cotton lint yield except at the Windhausen farm in 2009. In 2009, fumigation with the high rate of Telone resulted in a significantly lower lint yield than for any other treatments at both Windhausen farms. One possible explanation for this might be that these two fields were both non-irrigated. In 2009, there was considerably more rainfall throughout the growing season than in 2008. According to observations in the two management zones at Perryman farm, cotton grows better in soil that has higher moisture. Therefore, the excessive rainfall in 2009 may have provided the cotton plants with improved growing condition and thus reduced the damage *M. incognita* was able to cause, because *M. incognita* causes damage in plants only through magnifying the stress that the host plants encounter. When this stress is alleviated, the damage from *M. incognita* is also reduced. The lint yield in 2009 at both Windhausen farms was generally about 300 kg/ha more than that of 2008, which indicated the plants had better growth in 2009. Although plant vegetative growth was affected by nematicide treatments in 2009, cotton lint yield may be a less sensitive character to *M. incognita* infection. The reason we did not see the same results from Perryman farm in 2009 may because the soil at Windhausen farms had a better sustainability of moisture.

The fiber quality results showed no consistent and very little significant differences among treatments. Therefore, fiber quality may be less sensitive to *M. incognita* infection than

lint yield and more affected by other factors. For example, differences in fiber quality were not seen in 2008, although lint yield was significantly different among nematicide treatments. In the early stage of cotton growth, carbohydrates produced by leaves are mainly transported through the phloem to the roots, which act as the main carbohydrate sinks during this phase. In the boll development phase, the developing bolls become much stronger carbohydrate sinks than roots and shoots. Therefore at this stage, boll development dominates plant growth. This source to sink relationship in cotton also applies to inorganic nutrients and water (Ritchie et al., 2007). The mechanism of *M. incognita* parasitism is through manipulations of plant gene expression, instead of a direct interruption of plant cell formation of lesions on the roots (Williamson and Gleason, 2003). Therefore the defense reaction from plants is suppressed and the plants recognize *M. incognita* as strong metabolic sinks (McClure, 1977). The plants automatically transport nutrients to the nematodes' feeding sites. It is not clear whether *M. incognita* or the developing boll is a stronger nutrient sink during the boll development phase. If the developing bolls are stronger sinks, the nutrients will be transported preferentially to the developing bolls, the remainder of the nutrients being transported to nematode feeding sites. In this way, fiber development and fiber quality will not be affected. Another possibility is that *M. incognita* might be a stronger nutrient sink than developing bolls, or both are of equal priority for nutrient transport. In this situation, when sufficient nutrients exist, both developing bolls and *M. incognita* can still get sufficient nutrients, and fiber quality may not be affected either.

It is also possible that if *M. incognita* is not able to compete with the developing bolls for nutrients, they may, by manipulating plant genes, cause a reduction in the number of bolls allowing more nutrients to flow into their feeding sites. A reduced boll number by *M. incognita*

infection was documented in the greenhouse results from Chapter 2, and reduced yield by *M. incognita* infection was documented in the field trials.

From the results of the principal component analysis, *M. incognita* had negative effects on some fiber properties to include micronaire, staple, strength, fiber reflectance, fiber yellowness, length and uniformity. However, only the effect on micronaire was significant, and the findings were not consistent in the 2009 trial. Many of the fiber properties were also positively affected by nematodes. Therefore, it was not clear whether factors associated with *M. incognita* generally had positive or negative effect on overall cotton fiber quality, In the 2009 trials, the nematode effect was not as strong as in 2008 as only the sixth component contained information related to nematodes and the seventh component involved some nematode effect. Out of the 70.3% variation these seven principal components explained, these two components together only explained 8.21% of the total variation. This may be because in 2009 leaf nutrient contents were tested and incorporated into the principal component analysis. The leaf content may have provided more explanation for the variation. Also in 2009, excessive rainfall and excellent growing conditions may be a reason why the effect of nematodes was less obvious. Because the plants had better growing conditions, fiber development may have been less sensitive to *M. incognita* infection.

The values of fiber properties usually change in a very small range. However, a small change could bring the fiber quality below the USDA standard and leave the growers with penalties when selling their products. Therefore, although the difference in fiber properties among treatments was not statistically significant, it is still necessary to evaluate the actual price of cotton lint due to the changes in fiber quality associated with *M. incognita* infection. By evaluating the adjusted revenue, which is the total income deducted by the investment of crop

management and nematicide application, one can compare the economical profits that different treatments generated, which take into account both lint yield and fiber quality.

Different nematicide treatments did not alter the lint price significantly by affecting fiber quality. The average price of cotton fiber for 2008 and 2009 was $0.65/lb. The price of cotton fiber at Perryman and Windhausen farms was overall below the average price. None of the treatments improved the price to reach the average. Because of that, the adjusted revenue was usually significantly higher with a less expensive nematicide at Windhausen farms. At Perryman farm, the adjusted revenue was not affected by Telone treatments. The different results from Perryman and Windhausen fields might be due to their different growing conditions. In fields with better growing conditions, *M. incognita* is less likely to cause severe damage in cotton. Therefore with optimum cotton growing condition, the lowest investment for nematode control can result in the highest revenue. From the results of lint yield in 2009, which was presented in earlier in this chapter, we suspected that Windhausen West and Windhausen East fields may have better water sustainability than Perryman farm. If this was the case, then it supports the speculation that the reason adjusted revenue was the highest on the Windhausen farm with the least expensive nematicide control option was because cotton growing conditions were better there.

3.6 Summary and Conclusions

At three fields in both years, variable *M. incognita* population levels were successfully created with use different nematicide types. Fumigation with Telone II produced the lowest *M. incognita* population and use of seed treatments produced the highest nematode populations at the Perryman farm. Correspondingly, *M. incognita* infection assessed as gall ratings and

nematode reproduction (egg counts per gram of root) were both significantly lower with Telone II treatments and were the highest with seed treatments. Plant vegetative growth assessed as area under shoot height and root weight progress curves, height-to-node ratio, and shoot dry weight was usually the greatest where plots were fumigated with Telone II and reduced where seed treatments were used. Temik 15G provided better control of *M. incognita* than did seed treatments and therefore resulted in a better plant growth. Although sometimes the higher rate of a nematicide provided better numerical control, the differences between low and high rates of a specific nematicide were not significant in most cases.

Cotton lint yield in 2008 was significantly improved with use of Telone treatments in most cases, as a result of improved plant growth, although at Perryman farm, the improvement was not statistically significant. In 2009, fumigation with Telone II consistently resulted in greater yield at the Perryman farm, but appeared to result in a reduced yield in the two Windhausen fields. This may be because the excessive rainfall in 2009 provided a better growing condition for cotton, therefore, with less stress in plants, *M. incognita* damage was also suppressed. The fact that plant growth was affected by different nematicide treatments in 2009 suggested that cotton lint yield was less sensitive to *M. incognita* infection than plant growth, or perhaps was affect significantly by other factors as well.

The fiber quality results from all three fields in two growing seasons showed that in most cases, fiber quality was not consistently or significantly affected by nematicide treatments. Although in certain trials a few fiber properties differed significantly among treatments, these differences were not consistent across multiple trials. Principal component analysis provided a more direct relationship between fiber quality and *M. incognita* and other variables. These results showed that the effect of *M. incognita* on fiber quality was not significant in most cases and also

not consistent in different years. It was not clear from this whether the effect of nematodes on overall fiber quality was positive or negative. Fiber quality is likely less sensitive to *M. incognita* infection than lint yield. This may be because in the boll development phases of cotton growth, the developing bolls are nutrient sinks, as are *M. incognita* juveniles that are feeding in root tissues. If developing bolls are stronger sinks or if sufficient nutrient can be provided to the bolls despite the presence of *M. incognita*, fiber development and fiber quality will not be affected.

In most cases, there appeared to be no interactions between treatments and management zones. Nematode populations, plant growth, yield and fiber quality all responded the same to treatments within high- and low-risk management zones.

The risk management zone strategy has been implemented successfully in other fields to manage *M. incognita* in cotton (Ortiz, 2008). According to the risk management zone concept, growers can apply different types or rates of nematicides in order to obtain maximum revenue. In this study, management zone effects were not obvious at any field site in either year. Therefore, the risk management zone strategy may not be suitable for every field to control *M. incognita*s.

The results of the economical analyses from Perryman and Windhausen farms showed that cotton lint price was not affected by different treatments. The adjusted revenue was significantly higher in plots treated with Temik 15G than with Telone II in fields with better growing conditions or lower nematode populations.

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Table 3.1. Soil sampling dates for southern M. incognita juvenile counts at Perryman, Windhausen West and Windhausen East farms in 2008 and 2009.

Year	Field	Sampling date			
		First	Second	Third	Fourth
2008	Perryman	6/4/08	7/22/08	9/4/08	11/4/08
	Windhausen West	5/21/08	7/30/08	9/11/08	10/23/08
	Windhausen East	5/21/08	7/30/08	9/11/08	10/23/08
2009	Perryman	6/23/09	7/21/09	9/15/09	10/28/09
	Windhausen West	6/19/09	7/21/09	9/15/09	11/12/09
	Windhausen East	6/19/09	7/21/09	9/15/09	11/12/09

Table 3.2. Effect of nematicides on cotton root damage, *M. incognita* reproduction, and nematode juvenile population at Perryman farm in 2008 and 2009.

Year	Treatment	AUNPC[a] (day/100 cm^3)	AUGPC[b] (day)	AUEPC[c] (day/g)
2008	AERIS	39,643 a[d]	79.732 a	41,439 b
	AVICTA	38,365 a	89.889 a	75,406 a
	Low[e] Temik	40,913 a	57.596 b	30,355 bc
	High Temik	36,351 a	42.720 c	14,442 cd
	Low Telone	8,757 b	7.622 d	814 d
	High Telone	5,902 b	3.246 d	458 d
	P value	<.0001	<.0001	<.0001
2009	AERIS	31,972 ab	74.34 a	62,678 a
	AVICTA	38,400 a	60.70 a	38,930 ab
	Low Temik	30,030 ab	22.05 b	3,024 c
	High Temik	23,521 bc	28.14 b	6,018 bc
	Low Telone	13,063 cd	3.18 b	265 c
	High Telone	4,591 d	9.85 b	7,565 bc
	P value	0.0001	<.0001	0.0077

[a] Area under the nematode count progress curve. *M. incognita* juveniles were collected four times throughout the cotton growing season and were counted per 100 cm^3 soil.
[b] Area under the gall rating progress curve. Gall rating was based on a 0-5 index, 0 = no galling, 1 = trace infection with a few small galls, 2 = galling evident on <25% of the roots, 3 = 25-50%, 4 = 50-75% and 5 = >75% of the roots galled.
[c] Area under the egg count progress curve. Egg counts were *Meloidogyne* egg counts per gram of root.
[d] LSD(0.05) comparisons among treatments within a trial. Means in a column followed by the same letter are not significantly different. Data for two management zones was pooled when no significant interaction occurred.
[e] Low Temik = 3.36 kg/ha; High Temik = 6.72 kg/ha; Low Telone = 28.10 L/ha; High Telone = 56.12 L/ha.

Table 3.3. Effect of nematicides on cotton root damage, *M. incognita* reproduction, and nematode juvenile population at Windhausen farms in 2008 and 2009.

Year	Treatment	AUNPC[a] (day/100 cm^3)	AUGPC[b] (day)	AUEPC[c] (day/g)
2008	Low[e] Temik	21,534	34.497 a[d]	26,947
	High Temik	21,574	40.339 a	39,194
	Low Telone	17,732	7.339 b	9,750
	High Telone	10,646	1.872 b	1,480
	P value	0.0826	<.0001	<.0001
2009	Low Temik	60,867 a	58.806 a	25,865
	High Temik	60,752 a	46.571 a	11,194
	Low Telone	44,005 ab	6.838 b	2,856
	High Telone	30,944 b	17.963 b	22,926
	P value	0.0029	<.0001	0.2623

[a] Area under the nematode count progress curve. *M. incognita* juveniles were collected four times throughout the cotton growing season, and were counted per 100 cm^3 soil.
[b] Area under the gall rating progress curve. Gall rating was based on a 0-5 index, 0 = no galling, 1 = trace infection with a few small galls, 2 = galling evident on <25% of the roots, 3 = 25-50%, 4 = 50-75% and 5 = >75% of the roots galled.
[c] Area under the egg count progress curve. Egg counts were *Meloidogyne* egg counts per gram of root.
[d] LSD(0.05) comparisons among treatments within a trial. Means in a column followed by the same letter are not significantly different. Data for two management zones was pooled when no significant interaction occurred.
[e] Low Temik = 3.36 kg/ha; High Temik = 6.72 kg/ha; Low Telone = 28.10 L/ha; High Telone = 56.12 L/ha.

Table 3.4. Effect of nematicides on cotton shoot height, shoot dry weight, root weight, and height-to-node ratio at Perryman farm in 2008 and 2009.

Year	Treatment	AUHPC[a] (cm·day)	AURWPC[b] (g·day)	Height-to-node ratio (cm)	Dry weight (g)
				31DAP[c]	31 DAP
2008	AERIS	2,602.7 c[d]	509.67	2.56 cd	2.16 c
	AVICTA	2,668.4 c	521.88	2.49 d	1.97 c
	Low[e] Temik	2,782.5 bc	533.72	2.81 b	3.08 ab
	High Temik	2,733.7 c	529.59	2.71 bc	2.57 bc
	Low Telone	3,966.9 ab	555.46	3.03 a	3.68 a
	High Telone	3,181.4 a	625.40	3.06 a	3.66 a
	P value	<.0001	0.1909	<.0001	<.0001
				39 DAP	39 DAP
2009	AERIS	3,554.9	472.39	8.7 c	2.36 c
	AVICTA	3,535.4	485.15	9.3 bc	3.20 bc
	Low Temik	3,689.4	473.98	9.8 ab	3.93 ab
	High Temik	3,686.0	485.06	9.4 abc	3.85 ab
	Low Telone	3,812.1	542.91	10.1 ab	4.81 a
	High Telone	4,036.6	564.37	10.3 a	5.01 a
	P value	0.2315	0.5749	0.008	0.011

[a] Area under the height progress curve. Cotton shoot heights were measured twice in 2008 and four times in 2009 throughout the cotton growing season.
[b] Area under the root weight progress curve. Cotton root weights were measured twice in 2008 and 2009 throughout the cotton growing season.
[c] Days after planting
[d] LSD(0.05) comparisons among treatments within a trial. Means in a column followed by the same letter are not significantly different. Data for two management zones was pooled when no significant interaction occurred.
[e] Low Temik = 3.36 kg/ha; High Temik = 6.72 kg/ha; Low Telone = 28.10 L/ha; High Telone = 56.12 L/ha.

Table 3.5. Effect of nematicides on cotton shoot height, shoot dry weight, root weight, and height-to-node ratio at Windhausen farms in 2008 and 2009.

Year	Treatment	AUHPC[a] (cm·day)	Height-to-node ratio (cm)	AURWPC[b] (g·day)	Dry weight (g)
			43 DAP[c]		43 DAP
2008	Low[e] Temik	2,548.12	2.87	594.23	8.53
	High Temik	2,554.28	2.82	543.23	7.98
	Low Telone	2,583.17	2.89	558.00	7.89
	High Telone	3,696.62	2.86	573.61	8.29
	P value	0.1527	0.8841	0.2799	0.8580
			46 DAP		37 DAP
2009	Low Temik	2,719.30 b[d]	3.69	491.20	2.38 b
	High Temik	2,745.61 b	3.87	476.53	2.46 b
	Low Telone	2,927.33 a	3.86	530.74	2.85 a
	High Telone	2,995.04 a	3.86	537.43	2.68 ab
	P value	<.0001	0.4418	0.3548	0.0567

[a] Area under the height progress curve. Cotton shoot heights were measured twice in 2008 and four times in 2009 throughout the cotton growing season.
[b] Area under the root weight progress curve. Cotton root weights were measured twice in 2008 and 2009 throughout the cotton growing season.
[c] Days after planting
[d] LSD(0.05) comparisons among treatments within a trial. Means in a column followed by the same letter are not significantly different. Data for two management zones was pooled when no significant interaction occurred.
[e] Low Temik = 3.36 kg/ha; High Temik = 6.72 kg/ha; Low Telone = 28.10 L/ha; High Telone = 56.12 L/ha.

Table 3.6. Effect of nematicides on cotton yield and fiber quality at Perryman farm across all sampling plots in 2008 and 2009.

Year	Treatment	Color grade[a]	Staple (32nds)[b]	Micronaire	Strength (g/tex)	Leaf grade	Reflectance	Yellowness	Trash (%)[c]		Length (cm)	Uniformity (%)	Lint yield (kg/ha)
		---------- Pooled data[d] ----------							Zone 1[e]	Zone 2	---------- Pooled data ----------		
2008	AERIS	45	33.3	5.04	28.56	7	74.46	63.46	1.92 ab[f]	1.77	2.64	81.48	578.4
	AVICTA	49	33.8	5.04	28.49	7.42	73.85	63.79	2.06 a	1.89	2.64	80.9	524.5
	Low Temik[g]	45.44	33.11	5.04	28.08	5.75	74.43	63.13	1.58 bc	1.51	2.67	80.73	643.8
	High Temik	46.56	33.56	4.89	28.28	6.75	74.78	63.33	1.43 c	1.63	2.69	81.18	725.5
	Low Telone	46	32.9	5.15	28.41	6.58	74.97	64.42	1.88 ab	1.71	2.62	81.24	704.1
	High Telone	47	33.5	5.03	28.93	7.25	73.47	63.17	1.58 bc	1.83	2.64	81.23	770.7
	P value	0.5324	0.3502	0.5250	0.8265	0.2208	0.4032	0.6094	0.0389	0.4002	0.4351	0.6434	0.1365
		---------- Pooled data ----------						Zone 1	Zone 2		---------- Pooled data ----------		
2009	AERIS	46	37	4.65	30.75	6	73.9	63.67	67.33 a	1.25	2.92 abc	83.9	1004.84 b
	AVICTA	44.33	36.67	4.52	29.95	6	73.8	67	62 b	1.1833	2.98 a	82.87	991.02 b
	Low Temik	46	36.67	4.48	29.63	6.17	74.47	64.67	67.33 ab	1.2167	2.91 bc	83.73	1022.03 b
	High Temik	42.67	37	4.63	30.47	6.17	74.52	65	68 a	1.1667	2.94 ab	83.2	991.02 b
	Low Telone	42.67	36.67	4.67	29.93	5.33	74.55	66	68.33 a	1.15	2.87 c	83.05	1102.17 ab
	High Telone	41	37.33	4.53	30.52	6.5	74.88	65.67	67.67 a	1.1667	2.92 abc	83.32	1168.86 a
	P value	0.3637	0.6200	0.4430	0.2428	0.5408	0.7340	0.3804	0.0442	0.9918	0.0371	0.5687	0.0179

[a] Cotton fiber quality properties.
[b] Staple length (32nds of an inch).
[c] The percent of the sample surface covered by trash particles as determined by image analysis
[d] Data for two management zones was pooled when no significant interaction occurred.
[e] Zone 1 = low-risk zone; Zone 2 = high-risk zone.
[f] LSD(0.05) comparisons among treatments within a trial. Means in a column followed by the same letter are not significantly different.
[g] Low Temik = 3.36 kg/ha; High Temik = 6.72 kg/ha; Low Telone = 28.10 L/ha; High Telone = 56.12 L/ha.

Table 3.7. Effect of nematicides on cotton yield and fiber quality at Windhausen farms across all sampling plots in 2008 and 2009.

Year	Treatment	Color grade[a]	Staple (32nds)[b]	Micronaire	Strength (g/tex)	Leaf grade	Reflectance	Yellowness	Trash (%)[c]	Length (cm)	Uniformity (%)	Lint yield (kg/ha)
		---------- Pooled data[d] ----------										
2008	Low[g] Temik	44.68	35.95	4.37	30.13	6.95	75.22 a[e]	63.58	1.50 b	2.83	82.07	1119.6 b
	High Temik	45.00	35.75	4.33	29.87	6.90	74.68 ab	64.15	1.60 ab	2.82	81.76	1094.7 b
	Low Telone	45.00	36.05	4.35	30.56	6.85	74.53 ab	64.70	1.71 ab	2.85	82.21	1101.5 b
	High Telone	46.56	35.89	4.39	29.70	7.22	74.07 b	64.67	1.81 a	2.83	81.78	1207.1 a
	P value	0.673	0.719	0.890	0.073	0.184	0.035	0.419	0.036	0.327	0.226	0.047
		Zone 1[f]	Zone 2	Pooled data	Zone 1	Zone 2	Zone 1	Zone 2	Pooled data		Zone 1	Zone 2	------ Pooled data ------			
2009	Low Temik	47.67	42.67	36.92	4.55	4.17 b	29.62 a	29.28	6.83	75.04	60.17	64.17 a	1.38	2.92	83.06	1391.5 a
	High Temik	42.67	44.33	36.67	4.32	4.33 ab	29.80 a	28.65	6.83	75.03	63.17	62.17 a	1.42	2.91	83.00	1372.3 a
	Low Telone	44.33	44.33	36.42	4.30	4.52 a	28.37 b	28.93	6.67	75.16	63.33	61.33 ab	1.36	2.89	82.02	1335.7 a
	High Telone	41.00	47.67	37.08	4.28	4.58 a	29.28 a	29.78	6.42	75.03	65.33	59.17 b	1.18	2.94	82.66	1265.4 b
	P value	0.078	0.303	0.081	0.076	0.017	0.006	0.063	0.476	0.895	0.058	0.023	0.053	0.063	0.060	0.002

[a] Cotton fiber quality properties.
[b] Staple length (32nds of an inch).
[c] The percent of the sample surface covered by trash particles as determined by image analysis
[d] Data for two management zones was pooled when no significant interaction occurred.
[e] LSD(0.05) comparisons among treatments within a trial. Means in a column followed by the same letter are not significantly different.
[f] Zone 1 = low-risk zone; Zone 2 = high-risk zone.
[g] Low Temik = 3.36 kg/ha; High Temik = 6.72 kg/ha; Low Telone = 28.10 L/ha; High Telone = 56.12 L/ha.

Table 3.8. Effect of nematicides on cotton yield and fiber quality across the entire field at Perryman farm in 2008 and 2009.

Year	Treatment	Color grade[a]	Staple (32nds)[b]	Micronaire	Strength (g/tex)	Leaf grade	Reflectance	Yellowness	Trash (%)[c]	Length (cm)	Uniformity (%)	Lint yield (kg/ha)
2008	AERIS	29.333	32.8333	5.2333	28.5167	2.8333	81.0167	73.6667	0.16667	2.616	80.9333	505.6 b[d]
	AVICTA	29.333	33	5.2	29.2333	3	80.6333	73.6667	0.2	2.625	81.1	539.77 b
	Low[e] Temik	28.143	32.8571	5.1286	28.5286	3	81.1	73.7143	0.21429	2.609	80.6857	632.4 ab
	High Temik	31	32.8571	5.1571	28.9143	2.8571	80.8	73	0.17143	2.616	80.8143	628.02 ab
	Low Telone	28.143	33.1429	5.2286	28.8286	2.8571	80.8143	73	0.2	2.623	80.9714	692.28 a
	High Telone	28.143	32.8571	5.2429	28.5143	3	80.7714	72.2857	0.17143	2.609	81.0000	751.75 a
	P value	0.77	0.98	0.89	0.79	0.89	0.90	0.35	0.43	0.9909	0.74	0.0063
2009	AERIS	41 a	36.8	4.58	30.10 ab	2.8	77.02 b	72.6	0.32	2.885	82.3	965.2 b
	AVICTA	31 c	36.4	4.58	30.34 a	2.8	78.18 a	75.6	0.36	2.885	82.2	958.8 b
	Low Temik	33 bc	36.8	4.52	30.86 a	3	77.60 ab	76	0.32	2.911	82.34	981.3 b
	High Temik	33 bc	36.6	4.64	30.50 a	3	77.48 ab	76.4	0.4	2.891	82.18	976.0 b
	Low Telone	35 bc	36.8	4.66	29.88 ab	2.8	77.62 ab	75	0.3	2.880	81.96	1105.8 a
	High Telone	37 ab	36	4.78	28.98 b	2.8	77.42 b	73.6	0.32	2.850	81.72	1127.7 a
	P value	0.0107	0.1486	0.1344	0.0788	0.8439	0.0983	0.1536	0.8063	0.3041	0.3556	0.0081

[a] Cotton fiber quality properties.
[b] Staple length (32nds of an inch).
[c] The percent of the sample surface covered by trash particles as determined by image analysis
[d] LSD(0.05) comparisons among treatments within a trial. Means in a column followed by the same letter are not significantly different.
[e] Low Temik = 3.36 kg/ha; High Temik = 6.72 kg/ha; Low Telone = 28.10 L/ha; High Telone = 56.12 L/ha.

Table 3.9. Effect of nematicides on cotton yield and fiber quality across entire fields at Windhausen farms in 2008 and 2009.

Year	Treatment	Color grade[a]	Staple (32nds)[b]	Micronaire	Strength (g/tex)	Leaf grade	Reflectance	Yellowness	Trash (%)[c]	Length (cm)	Uniformity (%)	Lint yield (kg/ha)
2008	Low[e] Temik	27.84	35.37	4.46	29.94	3.00	81.17	74.32 ab[d]	0.22	2.80	81.67	978.65 b
	High Temik	29.00	35.35	4.49	30.38	2.95	80.92	74.00 b	0.23	2.79	81.47	991.63 b
	Low Telone	28.14	35.29	4.51	30.34	2.95	80.95	74.95 ab	0.21	2.79	81.48	1068.86 a
	High Telone	27.50	35.50	4.50	30.25	3.00	80.83	75.60 a	0.25	2.81	81.71	1075.77 a
	P value	0.7675	0.7381	0.6251	0.2525	0.7233	0.1937	0.0100	0.5937	0.7851	0.6141	<.0001
2009	Low Temik	33.5	36.25	4.45	29.05	3.00	78.80 a	71.75	0.25	2.87	81.03	1196.46
	High Temik	31.0	36.00	4.45	29.65	2.75	78.98 a	72.25	0.38	2.87	80.93	1175.99
	Low Telone	36.0	35.83	4.50	28.65	3.00	77.57 b	73.83	0.40	2.84	81.17	1196.46
	High Telone	38,5	35.50	4.68	28.38	3.00	78.51 a	69.75	0.30	2.82	81.40	1188.53
	P value	0.2531	0.3742	0.5523	0.0992	0.7930	0.0226	0.2889	0.3695	0.7237	0.3748	0.1988

[a] Cotton fiber quality properties.
[b] Staple length (32nds of an inch).
[c] The percent of the sample surface covered by trash particles as determined by image analysis
[d] LSD(0.05) comparisons among treatments within a trial. Means in a column followed by the same letter are not significantly different.
[e] Low Temik = 3.36 kg/ha; High Temik = 6.72 kg/ha; Low Telone = 28.10 L/ha; High Telone = 56.12 L/ha.

Table 3.10. Eigenvectors of principal components and explanatory variables for Perryman and Windhausen East and West farms in 2008.

Explanatory variable	Prin1	Prin2	Prin3	Prin4	Prin5
Shoot height 1st	**0.347**[a]	0.078	0.183	0.076	0.069
Shoot height 2nd	0.13	0.058	-0.185	**0.615**	-0.051
Number of nodes	**0.31**	0.122	**0.255**	-0.182	0.108
Height-to-node ratio	0.238	-0.045	-0.046	**0.431**	-0.034
Fresh weight	**0.345**	0.087	0.148	0.041	0.05
Dry weight	**0.337**	0.088	0.189	0.028	0.036
Root weight 1st	**0.335**	0.074	0.179	-0.089	0.159
Root weight 2nd	0.142	0.13	0.051	0.173	0.14
Root gall rating 1st	**-0.263**	**0.295**	0.174	0.083	0.202
Root gall rating 2nd	-0.237	**0.297**	0.175	0.128	0.185
Egg counts 1st	-0.208	**0.267**	0.221	-0.145	0.108
Egg counts 2nd	-0.228	0.236	0.103	0.162	0.173
Nematode counts 1st	0.058	0.099	**0.382**	-0.158	**-0.367**
Nematode counts 2nd	-0.109	**0.311**	0.166	**0.323**	0.062
Nematode counts 3rd	-0.179	0.244	0.09	0.009	**-0.309**
Nematode counts 4th	0.026	0.172	0.175	-0.094	**-0.538**
Soil pH	0.018	0.25	**-0.392**	-0.142	-0.096
Soil Ca	0.155	**0.4**	**-0.348**	-0.072	-0.066
Soil K	0.022	0.247	**-0.287**	0.037	-0.222
Soil P	0.041	0.115	-0.179	**-0.301**	**0.478**
Soil Mg	0.222	**0.377**	-0.227	-0.172	-0.011

[a] Eigenvectors with an absolute value >0.25 are bold-faced.

Table 3.11. Regression analysis of cotton yield and fiber properties to principal components for Perryman and Windhausen East and West farms in 2008.

Response variable	R^2	P	Parameter estimate[a]				
			Prin1[a]	Prin2	Prin3	Prin4	Prin5
Lint yield	0.6142	<.0001	79.955**	21.569*	50.717**	-28.715*	-22.794
Color grade	0.1076	0.0121	0.097	0.224	-0.629*	0.988**	0.177
staple	0.5522	<.0001	0.285**	0.135*	0.351**	-0.584**	-0.006
micronaire	0.5382	<.0001	-0.089**	-0.079**	-0.096**	0.066**	0.017
Strength	0.2321	<.0001	0.188**	0.006	0.261**	-0.397**	-0.027
Leaf grade	0.0562	0.1909	0.051	0.064	-0.070	0.084	0.015
Color reflectance	0.1395	0.0017	-0.018	-0.014	0.198*	-0.357**	0.139
Color yellowness	0.0879	0.0374	0.037	-0.157	0.335*	-0.328	-0.265
Trash	0.0588	0.1686	0.001	0.026	-0.047*	0.033	-0.016
Length	0.5407	<.0001	0.021**	0.009*	0.025**	-0.042**	-0.002
Uniformity	0.2408	<.0001	0.134**	0.016	0.109*	-0.258**	-0.003

[a] Principal components significant at $P = 0.05$ (*) or 0.01 (**).

Table 3.12. Eigenvectors of principal components and explanatory variables for Perryman and Windhausen East and West farms in 2008.

Explanatory variable	Prin1	Prin2	Prin3	Prin4	Prin5	Prin6	Prin7
Shoot height 1st	**0.270**	0.105	-0.107	0.028	0.120	0.193	0.034
Shoot height 2nd	0.195	0.094	-0.163	**0.269**	0.047	0.019	0.016
Shoot height 3rd	0.151	0.141	-0.137	**0.288**	0.058	-0.060	0.110
Number of nodes 1st	0.198	0.190	-0.147	-0.029	0.107	0.135	0.001
Number of nodes 2nd	-0.040	**0.261**	-0.156	0.243	-0.080	-0.169	0.003
Height-to-node 1st	0.121	0.204	0.097	-0.022	-0.002	-0.070	0.003
Height-to-node 2nd	0.193	-0.205	0.046	-0.064	0.128	0.189	-0.015
Fresh weight	**0.254**	0.141	-0.144	0.004	0.115	0.153	-0.035
Dry weight	**0.270**	0.117	-0.119	0.009	0.114	0.138	0.027
Root weight 1st	**0.283**	0.079	-0.105	-0.047	0.094	0.138	0.040
Root weight 2nd	0.059	0.036	-0.125	-0.006	0.120	0.041	-0.028
Nodes above white flower 1st	-0.037	0.211	-0.029	**0.255**	-0.094	-0.129	-0.219
Nodes above white flower 2nd	-0.206	0.152	-0.069	-0.086	0.000	-0.037	-0.146
Root gall rating 1st	-0.067	-0.150	0.249	0.036	0.128	0.059	0.187
Root gall rating 2nd	-0.151	-0.108	0.201	0.167	0.243	0.169	0.202
Egg counts 1st	-0.070	-0.126	0.133	0.088	0.214	-0.105	**0.393**
Egg counts 2nd	-0.130	0.012	0.166	0.198	0.050	0.232	0.050
Nematode counts 1st	-0.116	-0.082	-0.139	0.065	0.102	**0.281**	0.119
Nematode counts 2nd	-0.090	-0.035	0.015	0.212	-0.068	**0.300**	-0.245
Nematode counts 3rd	-0.156	0.030	0.101	0.199	0.006	0.244	**-0.271**
Nematode counts 4th	-0.090	0.034	0.053	**0.286**	0.057	0.225	-0.171
Leaf Ca	-0.046	0.118	0.245	0.023	0.118	-0.234	-0.008
Leaf K	0.051	0.148	0.103	0.102	0.191	-0.099	0.181
Leaf Mg	-0.094	-0.064	0.141	**0.251**	-0.135	-0.034	-0.110
Leaf N	**-0.259**	0.145	-0.085	-0.109	0.073	-0.032	-0.058
Leaf P	-0.180	0.122	-0.150	-0.187	-0.035	0.046	0.115
Leaf S	-0.111	-0.022	-0.017	-0.022	**0.289**	0.084	0.030
Leaf Al	0.097	-0.077	0.111	-0.012	**0.408**	-0.238	**-0.354**
Leaf B	0.226	-0.180	0.160	-0.071	0.051	-0.055	-0.041
Leaf Cu	-0.237	0.189	-0.139	-0.017	0.023	-0.073	0.012
Leaf Fe	-0.036	-0.051	0.037	-0.137	**0.477**	-0.198	**-0.375**
Leaf Mn	-0.223	-0.012	-0.223	0.023	0.187	-0.012	0.132
Leaf Na	0.117	-0.153	0.136	**0.269**	-0.192	-0.121	0.036
Leaf Zn	-0.104	0.089	-0.153	**-0.322**	-0.076	0.034	-0.021
Soill bc	0.056	0.204	0.036	0.231	0.086	-0.202	-0.005
Soil pH	0.018	0.184	**0.285**	-0.169	-0.068	0.238	-0.042
Water pH	0.018	0.184	**0.285**	-0.169	-0.068	0.238	-0.042

Soil Ca	-0.030	**0.288**	0.220	-0.052	-0.003	0.015	0.026
Soil K	-0.014	0.248	0.140	-0.049	0.198	0.157	0.033
Soil Mg	-0.086	**0.297**	0.179	-0.024	0.003	0.062	-0.078
Soil Mn	-0.214	0.096	-0.141	0.077	0.220	0.047	**0.262**
Soil P	0.166	0.125	0.237	-0.115	-0.082	-0.073	0.148
Soil Zn	0.053	0.231	0.167	0.001	-0.029	-0.167	0.227

[a] Eigenvectors with an absolute value >0.20 are bold-faced.

Table 3.13. Regression analysis of cotton yield and fiber properties to principal components for Perryman and Windhausen East and West farms in 2009.

Response variable	R^2	P	Parameter estimates[a]						
			Prin1[a]	Prin2	Prin3	Prin4	Prin5	Prin6	Prin7
Lint yield	0.6480	<.0001	-36.078**	27.621**	-22.212**	-2.084	13.001	15.802	3.538
Color grade	0.0484	0.8091	-0.047	0.237	-0.039	0.245	-0.339	-0.172	0.322
staple	0.1182	0.2195	0.007	-0.066*	0.019	0.052	0.064	-0.005	0.071
micronaire	0.2211	0.0087	0.025**	-0.021*	0.017	-0.029	-0.027	0.016	-0.022
Strength	0.2639	0.0016	0.115**	-0.105**	0.063	0.090	-0.030	0.040	0.104
Leaf grade	0.2642	0.0016	-0.096**	0.045	-0.063	0.138**	-0.040	0.109	0.062
Color reflectance	0.1690	0.0518	-0.120*	-0.026	-0.018	-0.167*	0.128	0.081	-0.148
Color yellowness	0.3370	<.0001	0.561**	-0.045	0.306*	0.161	-0.089	-0.329	0.520
Trash	0.1816	0.0345	-0.018	0.029**	0.006	0.032	-0.009	-0.010	0.024
Length	0.1073	0.2858	0.000	-0.004	0.003	0.006	0.001	0.001	0.005
Uniformity	0.1839	0.0320	0.077*	-0.085*	0.076	0.023	0.136	-0.062	0.108

[a]. Principal components significant at P = 0.05 (*) or 0.01 (**).

Table 3.14. Effect of different types and rates of nematicides on cotton lint price ($/lb) at Perryman and Windhausen West and Windhausen East fields in 2008 and 2009.

Treatment (acre^{-1})	PM[a] 08		PM 09		WW 08		WW 09		WE 08		WE 09	
	Zone1[b]	Zone2	Zone1	Zone2	Zone1	Zone2	Zone1	Zone2	Zone1	Zone2	Zone1	Zone2
AERIS	0.56	0.56	0.61	0.61	-	-	-	-	-	-	-	-
AVICTA	0.55	0.55	0.63	0.60	-	-	-	-	-	-	-	-
Low Temik	0.58	0.56	0.63	0.61	0.60	0.59	0.58	0.61	0.59	0.60	0.58	0.61
High Temik	0.59	0.56	0.62	0.62	0.57	0.60	0.59	0.59	0.58	0.57	0.60	0.59
Low Telone	0.56	0.56	0.62	0.59	0.59	0.60	0.61	0.57	0.59	0.60	0.58	0.60
High Telone	0.57	0.55	0.61	0.63	0.59	0.59	0.59	0.60	0.58	0.58	0.62	0.61
P value	0.28	0.94	0.98	0.53	0.54	0.90	0.12	0.63	0.75	0.28	0.22	0.43

[a] PM = Perryman; WW = Windhausen West; WE = Windhausen East.
[b] Zone 1 = low-risk zone; Zone 2 = high-risk zone.

Table 3.15. Effect of different types and rates of nematicides on adjusted revenue at Perryman and Windhausen West and East fields in 2008 and 2009.

Treatment (acre^{-1})	PM[a] 08		PM 09		WW08		WW09		WE08		WE09	
	Zone1[b]	Zone2	Zone1	Zone2	Zone1	Zone2	Zone1	Zone2	Zone1	Zone2	Zone1	Zone2
AERIS	227.3	149.5	421.6	451.0	-	-	-	-	-	-	-	-
AVICTA	127.7	182.2	400.6	452.3	-	-	-	-	-	-	-	-
Low Temik	300.3	183.8	464.9	456.4	510.3	511.8	585.5 a[c]	666.8 a	468.6	461.7	600.0 a	645.6 a
High Temik	297.0	231.2	405.6	463.4	444.9	444.0	579.4 a	624.8 a	482.2	426.4	601.2 a	588.2 ab
Low Telone	246.8	164.1	421.6	453.1	483.7	406.6	540.3 ab	509.6 b	491.4	348.6	554.9 a	555.6 b
High Telone	236.7	165.9	460.9	435.0	441.6	429.5	482.5 b	457.0 b	417.3	418.1	468.8 b	507.5 b
P value	0.1029	0.9502	0.7243	0.9951	0.4105	0.1252	0.0412	0.0025	0.1499	0.0786	0.0078	0.0405

[a] PM = Perryman; WW = Windhausen West; WE = Windhausen East.
[b] Zone 1 = low-risk zone; Zone 2 = high-risk zone.
[c] LSD(0.05) comparisons among treatments within a trial. Means in a column followed by the same letter are not significantly different.

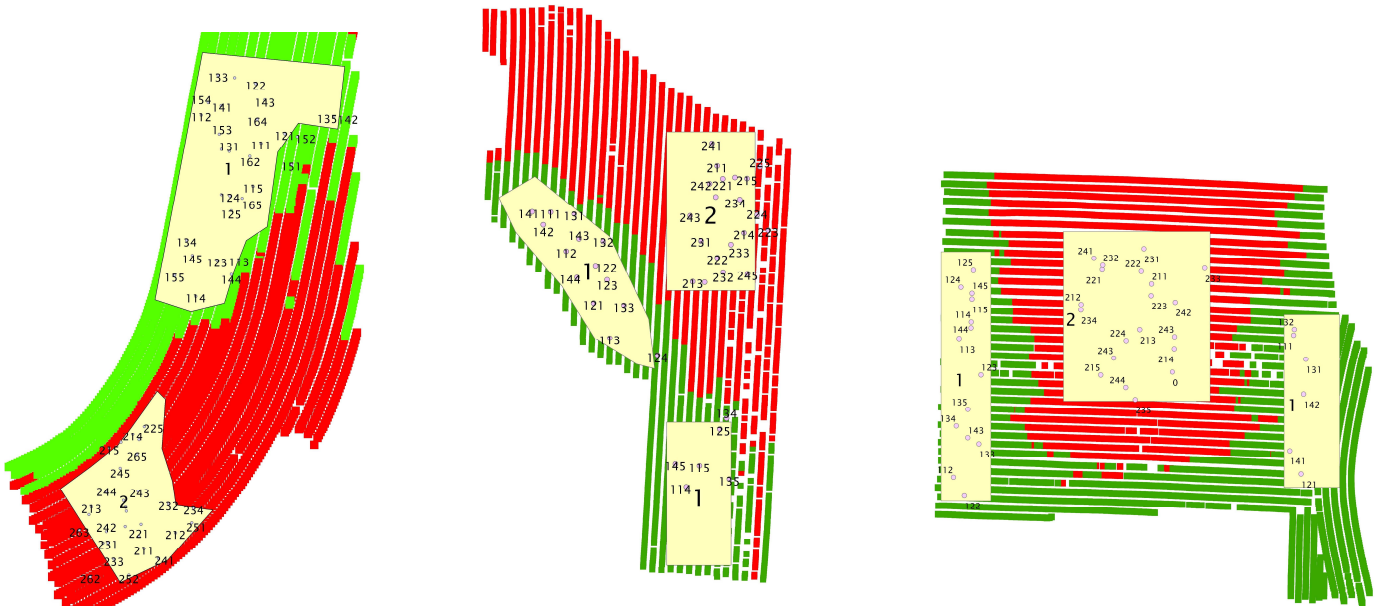

Figure 3.1 Experimental design maps of Perryman farm, Windhausen East farm and Windhausen West farm (from left to right) in 2008. Low- and high-risk zones are indicated by green and red coloring, respectively. Sampling plots are represented by 3-digit numbers, with the first digit corresponding to zone, the second digit corresponding to treatment, and the third number corresponding to replication. All sampling plots were completely randomized within each management zone, and treatments were applied in strips across the entire field.

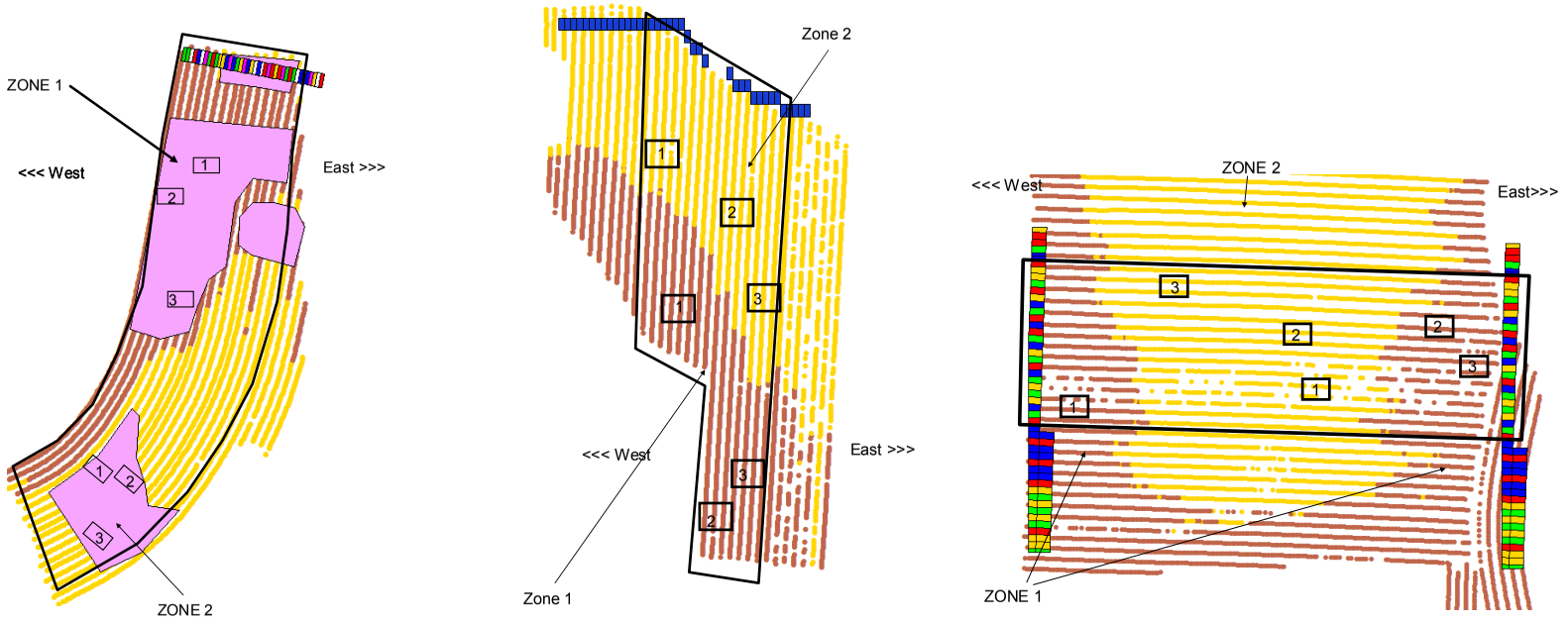

Figure 3.2 Experimental design maps of Perryman farm, Windhausen East farm and Windhausen West farm (from left to right) in 2009. Low- and high-risk zones are indicated by red and yellow coloring, respectively. Three sampling blocks were randomized within each management zone; treatments were randomized within each sampling block. Treatments were applied in strips across the entire field.

CHAPTER 4

EFFECT OF SOUTHERN ROOT KNOT NEMATODE (*MELOIDOGYNE INCOGNITA*) ON COTTON GROWTH, YIELD, AND FIBER QUALITY WITH DIFFERENT VARIETIES AND THE ECONOMIC IMPACT OF *M. INCOGNITA* ON COTTON PRODUCTION[1]

[1] Lu, P., Kemerait, R. C., Jr., Davis, R. F., and Perry, C. To be submitted to *Journal of Nematology*.

4.1 Abstract:

On two farms in southern Georgia, nematicides and cotton varieties were evaluated to further assess the impact of *Meloidogyne incognita* on cotton growth, yield, and fiber quality. The three chemical treatments used included Gaucho Grande seed treatment, Temik 15G and Telone II. The two varieties were FiberMax 9063 B2F and Stoneville 4554 B2RF. In both fields, populations of *M. incognita* were significantly reduced and plant growth was significantly improved in plots fumigated with Telone II. Cotton lint yield was increased in plots fumigated with Telone II at the Gibbs farm but not at the Nugent farm. Yields in plots treated with Temik 15G were usually not statistically different from yields obtained in plots where Gaucho Grande seed treatment was used. Most fiber quality properties were not significantly or consistently affected by nematicide treatments. Many fiber properties were significantly better in FiberMax 9063B2F than in Stoneville 4554 B2RF for both fields. The results of economical analysis showed that fumigation with Telone II can be more profitable in fields with higher *M. incognita* populations and an environment that is less than optimal for plant growth.

Keywords: Cotton, Economic impact, Fiber quality, *Meloidogyne incognita*, Southern root-knot nematode, Yield

4.2 Introduction

Cotton (*Gossypium hirsutum L.*), is an oilseed and fiber crop. It is grown in more than 70 countries and is the single most important fiber crop worldwide. No crop competes with it in the potential of value-added processing (Basra, 1999). Cotton is also one of the oldest cultivated crops. It has been associated with human activity since before recorded history.

The U.S. is the third-largest producer of cotton in the world. Cotton production in the U.S. has increased 66% in the past 40 years (Mitchell, 2009). In recent years, the U.S. has produced about 20% of the world's annual supply (Mitchell, 2009).

Root knot nematodes occur worldwide and attack a diversity of crops. They cause considerable losses of yield and affect the quality of the product, either by directly damaging plants (Kirkpatrick and Sasser, 1984), or by predisposing them to infection by fungal and bacterial pathogens (Powell, 1971).

The southern root knot nematode [*Meloidogyne incognita* (Kofoid & White) Chitwood] is found in all cotton production regions in the U.S. and is the most widely distributed nematode parasite of economic importance of the crop (Thomas and Kirkpatrick, 2001). It is considered the major yield-limiting plant-parasitic nematode across the U.S. Cotton Belt; approximately twice as much yield loss is attributed to *M. incognita* than to all other nematode parasites of the crop (Koenning et al., 2004). The estimated yield loss of cotton caused by *M. incognita* in the U.S. was 2.4% in 2007, which was greater than for any other cotton disease. This damage resulted in a loss of more than 106,000,000 kg of lint (Cotton Disease Loss Committee, 2008). In Georgia in 2007, *M. incognita* caused an estimated 6% reduction in yield resulting in a loss of 25,000,000 kg of lint (Cotton Disease Loss Committee, 2008).

Damage caused by plant-parasitic nematodes is distinct from other plant diseases because there are no unique symptoms on the above-ground portions of the plants. Nematodes are often unevenly distributed in the soil; therefore the symptoms associated with damage from nematodes may occur in irregular patches in the field (Beltwide Cotton Committee, 2003). These patches can be either small and limited in number, or large and widely distributed. Damaged plants may exhibit symptoms ranging from mild to severe stunting depending on the level of infestation, and a reduced rate of development. Foliage may also show symptoms of nutritional deficiency (Kirkpatrick et al. 1995). In the most severe cases, plants may die before maturation. The nematodes also interact with the Fusarium wilt pathogen (*Fusarium oxysporum* f.sp. *vasinfectum*), which leads to wilting and brown discoloration or necrosis of the vascular tissue of the lower stem (Beltwide Cotton Committee, 2003).

Below-ground symptoms caused by *M. incognita* on cotton can be much more diagnostic than above-ground symptoms. Visible galls or "knots" often appear on cotton roots (Bridge and Page, 1980). Swellings of the infected root tissue can be found on the cotton tap root and the lateral roots; however, the galls on cotton may not be as easy to observe as those on vegetable crops such as tomatoes. Galls are easier to detect if cotton plants are carefully dug (not pulled) from the soil. Also, the fine lateral roots need to be handled carefully when rinsed with water to remove soil (Beltwide Cotton Committee, 2003).

The tap root and its lateral roots are of vital importance to the cotton plant. A few galls on these roots can disrupt the normal flow of water and nutrients to the leaves and developing bolls, which can significantly reduce the yield of cotton (Bird and Loveys, 1975; Kirkpatrick et al., 1991; McClure, 1977).

Previous research has focused on the management of *M. incognita* to improve cotton yield. Very little data has documented the impact of *M. incognita* on cotton fiber quality. However, cotton fiber quality is a very important issue to the growers and to the textile industry. Fiber quality is a set of measurements that describe a sample of fibers extracted from a bale of cotton (Bradow and Davidonis, 2000). These measurements include length, uniformity, strength, micronaire, color grade, trash, leaf grade, preparation, and extraneous matter (USDA, 2001). They are compared with a set of standards from the U.S. Department of Agriculture (USDA) and are used to determine price premiums and discounts.

In recent years, cotton lint quality has become increasingly important because of the requirements from textile mills for fiber quality, the use of high-volume instrument (HVI) testing, the occurrence of discounts due to unfavorable fiber characteristics, and depressed cotton markets (Silvertooth, 1999). Georgia is the third-largest cotton producer in the U.S. In the 1990s and early 21st century, Georgia-produced cotton had the fiber quality second only to that produced in California, but now certain fiber quality parameters have not only fallen below levels found in western U.S cotton, but also below the average fiber quality of other southern and southeastern states (Bradow and Davidonis, 2000).

There are four general phases in cotton fiber development: 1) initiation, 2) elongation, 3) thickening, and 4) desiccation (maturation). Fiber length is determined in the elongation stage, which occurs about 21 days after flowering. During this stage, a thin cell wall of carbohydrate polymers is deposited allowing the fiber to elongate (DeLanghe, 1986). Water pressure inside the developing fiber has an influence on fiber elongation through regulating the deposition of carbohydrate polymers (Bradow and Davidonis, 2000). Therefore, if stresses occur during this stage (which would be typically associated with water stress or potassium deficiency), fiber

length can be reduced (Silvertooth, 1999). To some degree, fiber strength and uniformity can also be influenced by these same stresses (Bradow and Davidonis, 2000).

The thickening process of cotton fibers may overlap with elongation to some extent. During this stage, a series of carbohydrates produced through photosynthesis are deposited on the interior walls of the fiber, which increases the value of micronaire (Silvertooth, 1999). If this development is stopped prematurely, the micronaire value will decrease and finer fibers will occur. New bolls that are set on the plant usually have some of the greatest demand for carbohydrates. These bolls draw carbohydrates from the older bolls, which prevents the development of high-micronaire fibers on older bolls (Silvertooth, 1999).

The maturation of fiber occurs after the boll has opened and the metabolically inactive fibers dry. There is no quality measurement directly related to the maturation process. The fiber quality within a boll is at its utmost on the day of boll opening (Bradow and Davidonis, 2000). Therefore, harvesting should be as close to physiological maturity as possible to enhance the quality of the crop produced.

Environmental and management factors can significantly alter cotton fiber quality. For example, reduced light (cloudy) conditions result in the production of weaker fiber with reduced micronaire (Pettigrew, 2001). Early defoliation can also reduce micronaire in cotton (Snipes and Baskin, 1994).

Biological factors such as plant-pathogenic fungi and pests have also been studied. Cotton root rot, caused by the soilborne fungus *Phymatotrichum omnivorum*, can reduce fiber length and fineness significantly (Mulrean, 1984). It has also been well documented that by feeding on cotton bolls, stink bugs can decrease fiber quality as indicated by a reduction in almost all HVI-measured variables (Roberts et al., 2005). *Meloidogyne incognita*, a root-feeding

plant parasite, is another factor that could potentially affect fiber quality. *M. incognita* draws nutrients and water from cotton plants and exacerbates moisture, nutrient, or other kind of stresses and also fungal infection, thus indirectly causing damage to fiber development.

In previous studies, several fiber properties were documented to be affected by moisture and nutrient stress, reduced or delayed cotton growth, which in other studies, were associated with infection by *M. incognita*.

Fiber length and staple can be influenced by several factors including variety, temperature, water stress, nutrient deficiencies, and ginning practices (Bradow and Davidonis, 2000). Water relationships and irrigation practices have been studied primarily in relation to yield. One study conducted in the early 1980s indicated that fiber length was not impacted until water stress was such that yields were limited to less than 706.1 kg/ha (Grimes and Yamada, 1982). The interaction between *M. incognita* and water stress in cotton has also been studied. The results indicated that in susceptible cultivars, infection by *M. incognita* may decrease the movement of water from roots to leaves. The decrease in root flux caused by nematodes is equal to that induced by severe water deficit stress (Kirkpatrick et al., 1991). Other studies have demonstrated that water stress early in the bloom period had a less negative impact on fiber length than water stress late in the bloom period (Hearn, 1976; Marani and Amirav, 1971; Shimishi and Masani, 1971). Sensitivity of fiber elongation to severe water stress is apparently due to the physiological and mechanical processes of cell expansion (Hearn, 1994). As *M. incognita* infection gets more and more severe later in the season, the resulting water stress in the cotton plant may lead to a reduction in fiber length.

The possible impact of *M. incognita* on fiber strength is not as direct as the impact on fiber length. Firstly, fiber strength has a positive relationship with canopy sunlight absorption.

There is evidence that *M. incognita* can reduce plant leaf area, e.g., in tomato (Loveys and Bird, 1973). Therefore by reducing cotton leaf area, *M. incognita* may affect fiber strength. Secondly, studies have indicated that heat accumulation during the flowering period can also affect fiber strength. Fiber strength was greatest in bolls that developed from flowers produced during the first 4 to 6 weeks of flowering. Flowers that opened during the latter 2 weeks of the flowering period produced bolls with the lowest fiber strength (Jones and Wells, 1997). Therefore, *M. incognita* may reduce fiber strength by delaying cotton development and flowering.

Authors in previous studies have suggested several factors that could influence fiber micronaire. Significant differences in micronaire among commercially available varieties have been examined. A fiber thickens as cellulose is deposited inside the fiber cell. Cellulose is derived from photosynthetic carbon fixation. As fiber is thickened, the micronaire value increases. If photosynthesis of cotton is affected by *M. incognita*, as confirmed in tomato plants (Loveys and Bird, 1973), the lack of carbohydrate production may lead to a lower micronaire value. However, a micronaire value that is too high can also result in loss of profit. It is important for cotton to produce sufficient bolls later in the season to either compete for carbohydrates or to produce sufficiently lower micronaire fiber. This fiber can be blended with the higher micronaire fiber to reduce overall readings (Silvertooth, 1999). Therefore, the reduced growth and boll setting in cotton caused by *M. incognita* (Kirkpatrick et al., 1995; Walker et al., 1998) may affect the blended micronaire value.

As discussed before, water stress can be caused by *M. incognita* (Kirkpatrick et al., 1991). In other studies, it has also been shown that water deficiency can affect some properties of cotton fiber. Moisture stress later in the season was found to reduce fiber length and micronaire (Marani and Amirav, 1971). A reduction in fiber length due to water stress was also observed in a

subsequent study. Although other characters were not consistently affected, the distribution of bolls on the cotton plant was consistently and significantly affected by irrigation (Pettigrew, 2004).

Studies on the effect of *M. incognita* on nutrient concentrations in plants show that a change in concentration of the nutrients is likely one of the first effects of the nematode on host physiology. These changes in nutrient concentration alter host metabolism and contribute directly or indirectly to the chlorosis and premature leaf abscission on soybean plants (Melakeberhan et al., 1987). This is also true according to another study in soybean. The uptake of nitrogen, phosphorus, and calcium was affected by *M. incognita* (Carneiro et al., 2002). Many nutrients also have effects on fiber quality. For example, leaf nitrogen during the boll maturation period had significant positive correlations with fiber length and negative correlations with micronaire (Reddy et al., 2004). Potassium is also a very important element to cotton and has positive correlations with fiber length, micronaire index, fiber strength, and fiber length uniformity ratio (Cassman et al., 1990).

Approaches to effective nematode control include crop rotation, field sanitation, cover crops, varietal resistance, and nematicides. Nematode population densities can be reduced by selecting rotation crops that are not hosts to *M. incognita*, or by planting resistant cultivars (Rich and Kinloch, 2005). To date, no *M. incognita*-resistant cultivar is commercially available. Several cotton cultivars have been reported to have some level of tolerance to the Fusarium wilt/*M. incognita* disease complex. However, these cultivars do not show significant resistance to the nematodes, although some are resistant to Fusarium wilt (Beltwide Cotton Committee, 2003). Biological approaches for nematode control have been studied but have not been thoroughly explored in cotton production systems (Starr et al., 2007).

Chemical nematicides are used widely to control *M. incognita*. Three nematicide management strategies are currently applied widely in the U.S. (Koenning et al., 2004), including the application of aldicarb at rate of 0.8-1.2 kg/ha in the planting furrow; preplant soil fumigation using either 1,3-dichloropropene or metam-sodium (Starr et al., 2007); and the supplemental use of either aldicarb applied as a side-dress during the first third of the season or a foliar application of the carbamate oxamyl (Lawrence and Mclean, 2000, 2002). Out of all these strategies, at-planting application of aldicarb is perhaps the most universal nematicide strategy in the U.S. Aldicarb is applied on 20 to 30% of the cotton hectarage each year (Koenning, et al., 2004). In severely infested fields, nematicides can reduce the nematode population by more than 50% (Beltwide Cotton Committee, 2003). Using chemical nematicides can provide cotton with a zone of protected soil in which roots can develop for 4 to 6 weeks with reduced damage from nematodes. By protecting the crop during early development, yield losses can be reduced substantially even though nematodes may penetrate the roots during the latter part of the season (Beltwide Cotton Committee, 2003). Seed treatments have shown some promising results in protecting emerging roots from nematode infection (Monfort et al., 2006), but may not be sufficient in fields with high nematode population densities.

Greenhouse and field tests from previous chapters showed that *M. incognita* infection significantly reduced cotton growth, certain physiological characters, and cotton lint yield. Fiber quality was not significantly affected by *M. incognita* infection. Risk management zones did not have significant effects or had significant interactions with nematicide treatments, which had successfully created different levels of *M. incognita* infection with different types of nematicides. However, in the study in Chapter 3, all fields were planted with one variety, PD 555. This variety has no reported *M. incognita* resistance. In greenhouse test, however, resistant varieties had

better vegetative and reproductive growth, and resulted in significantly lower *M. incognita* infection and reproduction on cotton roots. Therefore, it is valuable to quantify whether resistance to *M. incognita* in cotton can lead to higher lint yield and fiber quality.

The objective of this chapter was to determine whether the effects of *M. incognita* infection on cotton growth, yield, and fiber quality are different in two cotton cultivars that have different levels of resistance to the pathogen.

4.3 Materials and Methods

The experiments were conducted at the Nugent farm in Coffee County, GA in 2008 and at the Gibbs farm in Tift County, GA in 2009. These two fields were both under irrigation and had histories of losses to *M. incognita*. The tillage method used at Nugent farm was conservation tillage (strip-tillage into a killed rye cover crop), whereas conventional tillage (ripped and bedded) was used at the Gibbs farm.

Two different varieties of cotton were planted, FiberMax 9063B2F and Stoneville 4554B2RF. FiberMax is highly susceptible to *M. incognita* infection, whereas Stoneville has some level of tolerance to the *M. incognita*/ Fusarium wilt complex (Barfield, 2003; Phipps and Eisenback, 2005). Three treatments were assessed to include: (T1) Gaucho Grande seed-applied insecticide (1-[(6-Chloro-3-pyridinyl)methyl]-N-nitro-2-imidazolidinimine at 0.375 mg ai/ seed); (T2) Temik 15G (aldicarb 6.72 kg/ha); and (T3) Telone II (1,3-dichloropropene 56.12 L/ha) + Temik 15G (aldicarb 3.36 kg/ha). The 3lb/a Temik treatment in T3 was used along with Telone II to provide thrips control. The decision to use these three treatments was based on the need to establish different populations of nematodes in the field. Gaucho Grande would not affect the nematode populations; Temik 15G at 6.72 kg/ha would offer some control; Telone II at 56.12

L/ha is too expensive for use in commercial cotton fields but should nearly eliminate nematodes from a plot. All treatments were applied in strips across the testing fields.

The experiment was conducted implementing a split-plot design at both field sites, with the main plot factor being cotton varieties and the sub-plot factor being nematicide treatments. There were four replications at the Nugent farm in 2008 and six replications at the Gibbs farm in 2009. The sampling plots were four rows by 15.2 meters in both fields, Telone II was applied prior to planting and Temik 15G was applied at planting along with herbicide.

M. incognita populations were determined by collecting soil samples from the center two rows of the sampling plots using soil probes. The soil samples were then sent to the University of Georgia's Nematology Lab in Athens for processing. Nematodes were extracted from 100 cm^3 soil using centrifugal-flotation (Jenkins, 1964) and *Meloidogyne* juveniles were counted under a dissecting microscope. Soil samples were taken three to four times during the season on a monthly basis (Table 4.1).

Cotton shoot height was recorded approximately every 2 weeks during the first half of the season and until the cotton plants reached "cut-out". From each sampling plot, five plants were randomly selected and measured. At approximately 4 and 8 weeks after planting, plant samples were taken from each field for further measurements. Five random plants from the center two rows of the sampling plots were carefully dug from the soil. The plants were divided into shoots and roots and these were stored in plastic sampling bags.

The measurements taken at first destructive sampling included the following: (1) Height-to-node ratio: Height-to-node ratio was calculated by dividing shoot height by the number of nodes. The number of nodes was recorded by counting the number of leaves that are bigger than a U.S. quarter coin above the cotyledons on the main stem. (2) Shoot dry weight: The five plant

tops from each sampling plot were packed into one paper bag and dried in an oven at about 60˚C for 2 days before dry weight was measured. (3) Root gall rating: Based on a 0-5 index, 0 = no galling, 1 = trace infection with a few small galls, 2 = galling evident on <25% of the roots, 3 = 25-50%, 4 = 50-75% and 5 = >75% of the roots galled (Kinloch, 1990). (4) Egg counts: Eggs of *M. incognita* were extracted with 0.625% NaOCl from cotton roots by shaking for 3 min. (Hussey and Barker, 1973) and counted microscopically.

For the second destructive sampling, only the cotton roots (n = 5) were sampled, because the tops were too large to handle. Therefore, root weight, gall rating and egg counts were measured at the second destructive sampling.

Data for variables that were measured repeatedly were transformed into areas under the variable progress curves using the trapezoidal method. The formula used to calculate the areas was the same as the formula for area under disease progress curve (AUDPC), which is $\text{AUDPC} = \sum_{i=1}^{n-1}\left(\frac{y_i + y_{i+1}}{2}\right)\left(t_{i+1} - t_i\right)$. Here, t is the time in days after planting at each measurement, y is the reading of the variable that was being measured, and n is the number of readings. The areas under the progress curve from each treatment were than analyzed using ANOVA and LSD tests in SAS (SAS Institute, Cary, NC) using the PROC GLM procedure.

At the Gibbs farm in the 2009 growing season, cotton leaves from the fifth node on the main stem were collected and sent for nutrient analysis to the University of Georgia's Soil Testing Lab in Athens. At harvest in 2008, the spatial variability of cotton yield (seed cotton) was recorded using an Ag Leader cotton yield monitor system (Ag Leader Technology, Ames, IA) installed on a Model 9965 four-row John Deere picker (John Deere, Moline, IL). In the 2008 study at the Nugent farm, yield was collected from the entire treatment strips. Cotton samples were also collected from across the entire treatment strips for fiber quality analysis. At the Gibbs

farm in 2009, cotton yield was determined by weighing the cotton sample collected by the picker from two rows of each sampling plot. These samples were also used for fiber quality tests. For both of these two fields, the cotton samples were ginned at the University of Georgia microgin. Cotton lint was then sent to the USDA cotton classifying office in Macon, GA for quality tests.

Results were analyzed with PROC GLM in SAS using ANOVA and LSD tests. The interaction between variety and Nematicide treatments was also analyzed. Principal component analysis was conducted in SAS using PROC PRINCOMP to elucidate the relation between response variables (yield and fiber quality) and explanatory variables (plant growth, nematode population and reproduction, soil nutrient contents, etc.). The principal components were interpreted by finding the explanatory variables with the highest absolute values of eigenvectors, usually above 0.25 to 0.30. Regression analysis of the most important principal components, which together explain over 70% of the total variation, was conducted against the response variables.

To evaluate the economic impact of *M. incognita* on cotton production relative to fiber quality, the lint price per pound was calculated using the Southeast base price from the spot cash market (averaged December 2008 and 2009 data) plus the premiums and/or minus the discounts for quality. The average base price equaled $0.6549/lb. In 2008 the price included a loan deficiency payment because it was below the loan rate of $0.52/lb. In addition to lint price, the adjusted revenue was also evaluated. The adjusted revenue took many important economical influencers into account, including nematicide application, variety, tillage, labor, yield, fiber quality. Both lint price and adjusted revenue were analyzed with SAS GLM using ANOVA and LSD tests.

4.4 Results

4.4.1 Plant growth, yield, and fiber quality

On the Nugent farm in 2008, three treatments were applied including Gaucho Grande seed treatment, 6.72 kg/ha Temik, and 56.12 L/ha Telone. *M. incognita* juvenile counts were the lowest where plots were fumigated with Telone II and he highest where plots were planted with seed treated with Gaucho Grande (Table 4.2). The difference between plots planted with seed treated with Gaucho Grande and plots treated with Temik 15G was not significant. At the Gibbs farm there was a statistical interaction between cotton varieties and nematicide treatments. Nematode counts were significantly higher in plots planted with variety FiberMax 9630B2F than in plots planted with Stoneville 4554B2RF, and there was a greater reduction in nematode counts with Telone treatment in FiberMax 9630B2F. However, the nematicide treatment effect was not statistically significant in either variety in this trial. In both trials, gall ratings and egg counts were the lowest with Telone treatment, but gall ratings were only significantly different among treatments in the Gibbs 2009 trial, and egg counts were only statistically separated between Gaucho and Telone in the Nugent 2008 trial. In most cases, plots planted to FiberMax 9630B2F resulted in higher nematode counts, higher gall ratings and higher egg counts than those planted to Stoneville 4554 B2RF, but only nematode counts in the Gibbs 2009 trial differed significantly.

In both trials, cotton shoot height and root weight were significantly higher in plots fumigated with Telone II. Height-to-node ratio and shoot dry weight were statistically greater in plots fumigated with Telone in the Gibbs 2009 trial. There was no interaction between varieties and nematicide treatments relative to cotton growth. The variety effect was only significant for shoot dry weight in the Nugent 2008 trial, where FiberMax 9630B2RF had a significantly higher shoot dry weight. However, it was reverse in the Gibbs 2009 trial.

Cotton lint yield was the greatest in plots fumigated with Telone II in both trials (Table 4.4), but the yield was only significantly higher in the Gibbs 2009 trial. There was no significant difference between Gaucho Grande and Temik treatments in yield in either trial. The variety effect on lint yield was not significant in either trial (Table 4.5), and there was no interaction between varieties and nematicide treatments.

Most of the fiber quality properties did not differ significantly among treatments (Table 4.4). In the Nugent 2008 trial, the nematicide treatment effect was only significant on color grade, fiber reflectance and trash content. The highest color grade, fiber reflectance and trash content were obtained in lint from plots treated with Temik, Telone, and Temik respectively, and the lowest were from Telone, Temik, and Gaucho, respectively. There was no statistical interaction between variety and nematicide treatment for any of the fiber properties in the Nugent 2008 trial.

In the Gibbs 2009 trial, the only fiber property significantly affected by nematicide treatment was trash content. Plots planted to seed treated with Gaucho Grande resulted in significantly higher trash content than the other two treatments. There was a statistical interaction relative to micronaire between varieties and treatments. In FiberMax 9630B2F, micronaire values were greatest in lint from plots planted to seed treated with Gaucho Grande, whereas in Stoneville 4554B2RF, micronaire values were greatest in plots fumigated with Telone II. However, the treatment effect was not significant in either variety,

The difference between varieties relative to fiber quality was significant in many cases (Table 4.5). In both trials, staple, strength, fiber reflectance and fiber length were significantly higher in FiberMax 9630B2F. Fiber yellowness was significantly lower and uniformity was significantly higher in FiberMax 9630B2F only in the Gibbs 2009 trial.

4.4.2 Principal component analysis

In the Nugent trial conducted in 2008 (Table 4.6), the variables that gave the greatest contribution to principal component 1 were all plant growth variables. Therefore, principal component 1 was considered a plant growth variable. For principal component 2, the variables that contributed the most were all related to nematode factors, such as gall rating and nematode juvenile counts. Therefore, principal component 2 was considered a nematode component. Principal component 3 had both plant growth and nematode contributors and principal component 4 was again a nematode component. These four principal components together explained 75% of the variation.

In the regression analysis of cotton yield and fiber properties to the first four principal components (Table 4.7), the nematode component had a negative effect on yield, although the effect was not statistically significant. Principal components 1, 3, and 4 all had significant positive effect on cotton lint yield. Fiber strength, leaf grade, reflectance, yellowness, trash, length and uniformity all had a negative relationship with either principal component 2 or component 4, which were both nematode components. However, none of these relations were significant. Most fiber properties were mainly positively affected by plant growth.

Based on the eigenvectors from the principal component analysis for the Gibbs 2009 trial (Table 4.8), principal component 1 was a plant growth component, principal component 2, 3 and 5 were all nematode and leaf nutrient components, and principal component 4 was a leaf nutrient component. These first five principal components together explained 72% of the total variation. Regression of yield and fiber quality to principal components (Table 4.9) showed that the plant growth component significantly affected yield and fiber yellowness. Principal component 2 and 3, which were nematode plus leaf nutrient components, both had negative effects on yield and

almost all fiber properties. These two nematode plus leaf nutrient components explained 24% of the variation. However, only the effect of principal component 2 on lint yield and trash content was significant. Many of the fiber properties were also significantly affected by principal component 4, which was a leaf nutrient component.

4.4.3 Economic impact

In the two trials conducted here (Table 4.10), only the lint price of Stoneville 4554B2RF from the Gibbs 2009 trial significantly differed among treatments. Cotton harvested from plots treated with Temik and Telone had significantly improved lint prices compared with cotton from plots planted to the Gaucho Grande seed treatment. This was due to a higher efficiency of nematode control. For the Nugent 2008 trial, the adjusted revenue was the highest with seed treatment and the lowest with Telone treatment for FiberMax 9063B2F. In 2008, the Nugent field had the lowest nematode populations out of all five field sites assessed in this thesis. However, in the Gibbs 2009 trial, the adjusted revenue was the highest for cotton yields obtained from plots fumigated with Telone for both varieties.

4.5 Discussion

The results from the Nugent farm 2008 trial and the Gibbs farm 2009 trial showed that with different types of nematicides, different levels of nematode populations, infection, and reproduction were created successfully. Nematode pressure was higher in the Gibbs farm trial than in the Nugent farm trial. The greatest nematode populations, disease severity, and reproduction were found in plots planted to seed with Gaucho Grande seed treatment, and the lowest was from plots fumigated with Telone II.

Both trials showed that cotton plants were less stunted with better *M. incognita* control, as shoot height was greater in plots fumigated with Telone II. There was also a better root growth with more aggressive nematode control in both trials. However, comparing these two trials, the difference among treatments in plant growth was more obvious in the Gibbs 2009 trial. This trial showed that with better nematode control, cotton plants were less stressed as assessed by the higher height-to-node ratios.

The difference in shoot dry weight between the two cotton varieties in the Nugent 2008 trial may be due to their inherent genetic potential rather than different susceptibility to *M. incognita*, because no significant differences in *M. incognita* populations and disease severity were found between the two varieties. This variety effect was similar to the greenhouse results in Chapter 2.

Better plant growth and less stress have resulted in a higher lint yield in both trials, although for one of the trials the difference in yield was not significant. *M. incognita* not only stunted cotton plant growth, but also reduced cotton lint yield. Since there was no difference in nematode-associated disease severity between the two varieties, cotton lint yield also did not differ between varieties.

In both trials, most of the fiber properties were not significantly different with different treatments, except for one or two fiber properties.H However, the way these fiber quality properties differ among treatments did not follow the pattern of nematode populations, disease severity, or reproduction level. Therefore, these differences may be due to factors other than *M. incognita* infection. Also, none of the treatment effects on these fiber properties was consistent in any of the other trials (Chapter 3). In both of the current trials, many of the fiber properties were significantly different between FiberMax 9063B2F and Stoneville 4554B2RF. Many fiber

properties including staple length, strength, reflectance, trash content, and uniformity were all significantly higher with FiberMax 9063B2F than Stoneville 4554B2RF. From the results of the greenhouse trials in Chapter 2, Stoneville had a better tolerance of *M. incognita* than FiberMax, and none of the field test results showed *M. incognita* has affected fiber quality. Therefore the difference in fiber properties between these two varieties may be mainly due to their genetic differences.

The principal component analyses for these two trials showed that plant growth had a major impact on cotton yield and fiber quality. In the Nugent 2008 trial, many fiber properties were negatively affected by *M. incognita*, although the effect was not significant. In the Gibbs 2009 trial, the effect of *M. incognita* was confounded with leaf nutrient contents. This combined effect was usually not significant. However, the effect of leaf nutrients alone was usually significant. This may indicate that nematode variables and leaf nutrients were negatively correlated.

The values of fiber properties usually change in a very small range. However, even a small change in values could have brought the fiber quality below the USDA standard and leave the growers with penalties when selling their products. Therefore, although the difference of fiber properties among treatments was not statistically significant, it is still necessary to evaluate the actual price of cotton lint due to the changes in fiber quality associated with *M. incognita* infection. By evaluating the adjusted revenue, which is the total income reduced by the cost of crop management and nematicide application, we can compare the economic profits for the different treatments, which take into account both lint yield and fiber quality.

Nematode control at the Nugent farm did not significantly increase economic return, because nematode populations and disease severity were relatively low in this field. A greater

investment resulted in lower revenue. However in the Gibbs trial, the adjusted revenue was the highest with Telone treatment for both varieties. In this trial, both Temik and Telone treatments increased the lint price by 0.01$/lb compared with Gaucho seed treatment. From previous results, the Gibbs field had a high *M. incognita* pressure and also growing conditions that were not optimum for cotton growth, which presumably allowed *M. incognita* to cause more severe damage. That might be the reason why Telone was more beneficial in this trial than any other trial.

4.6 Summary and Conclusions

In both the Nugent farm 2008 trial and the Gibbs farm 2009 trial, different populations of *M. incognita* were created using three chemical treatments. With more aggressive nematode control, plant growth was significantly improved in terms of greater shoot height, height-to-node ratio, dry weight, and root weight. Significant differences were found mainly between Telone II and the other two treatments. Gaucho Grande and Temik treatments did not differ except for root gall rating. Better plant growth with Telone was reflected in a greater lint yield in this treatment. Although higher populations of *M. incognita* were associated with FiberMax 9630B2F, this was not found to affect plant growth or yield, indicating genetic characters played a greater role in plant growth.

Fiber quality was not affected by nematicide treatments significantly or consistently. Although few differences were observed in nematode populations, plant growth or yield between the two varieties, most fiber properties were significantly higher in the FiberMax variety in both the Nugent 2008 and Gibbs 2009 trials, This may be due to genetic characteristics rather than *M. incognita* infection, as Stoneville has a better tolerance of *M. incognita* infection than FiberMax

according to greenhouse results, and none of the field test results showed *M. incognita* affected fiber quality.

Since there were very few interactions between cotton varieties and nematicide treatments, we can conclude that the growth, yield, and fiber quality of the two varieties with different levels of resistance to *M. incognita* responded similarly to *M. incognita* infection. Different types of nematicides had the same effect on FiberMax 9630B2F and Stoneville 4554B2RF.

The principal component analysis for the Nugent 2008 trial showed that plant growth had a positive effect on most fiber properties while *M. incognita* had some level of negative effect on fiber quality, which was consistent with other trials in 2008. The results of principal component analysis for the Gibbs 2009 trial showed that the nematode effect may have been obscured by leaf nutrient factors because they may have been negatively correlated with each other.

The results of economic analysis of these two trials showed that cotton lint price was not affected by the different treatments. The adjusted revenue was significantly higher with Temik than with Telone in the field with better growing conditions or less nematode population. Telone application was much more profitable in the field with higher *M. incognita* pressure and an environment that was not optimum for plant growth.

4.7 References

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Table 4.1. Soil sampling dates for southern M. incognita juvenile counts at Nugent farm in 2008 and at Gibbs farm in 2009.

Trial	Sampling date		
	First	Second	Third
Nugent 2008	6/12/08	8/12/08	11/10/08
Gibbs 2009	7/24/09	9/17/09	12/7/09

Table 4.2. Effect of nematicides and cotton varieties on root damage, *M. incognita* reproduction and nematode juvenile population at Nugent farm in 2008 and Gibbs farm in 2009.

Field	Treatment	AUNPC[a] (day/100 cm^3)		AUGPC[b] (day)		AUEPC[c] (day/g)	
Nugent	Gaucho	24,352 a[d]		16.169		5670 a	
	Temik	21,954 a		18.548		3946 ab	
	Telone	4,539 b		8.905		766 b	
	P value	0.0395		0.0997		0.0338	
		FM	ST				
Gibbs	Gaucho	36,297	12,407	43.61 a		2694	
	Temik	28,907	15,502	16.18 b		602	
	Telone	6,777	2,301	2.33 c		240	
	P value	0.1197	0.3226	<.0001		0.2885	
Cultivar effect		FM[e]	ST	FM	ST	FM	ST
2008	Average	23993.65	9903.31	18.66	10.42	4429.90	2491.21
	P value	0.1522		0.3641		0.3506	
2009	Average	49485.29	9655.46	22.84	18.58	604.30	1752.60
	P value	0.0144		0.4012		0.4223	

[a] Area under the nematode count progress curve. *M. incognita* juveniles were collected four times throughout the cotton growing season, and were counted per 100 cm^3 soil.
[b] Area under the gall rating progress curve. Gall rating was based on a 0-5 index, 0 = no galling, 1 = trace infection with a few small galls, 2 = galling evident on <25% of the roots, 3 = 25-50%, 4 = 50-75% and 5 = >75% of the roots galled.
[c] Area under the egg count progress curve. Egg counts were *Meloidogyne* egg counts per gram of root.
[d] LSD(0.05) comparisons among treatments within a trial. Means in a column followed by the same letter are not significantly different.
[e] FM = FiberMax 9630B2F; ST = 4554B2RF.

Table 4.3. Effect of nematicides and cotton varieties on shoot height, shoot dry weight, root weight and height-to-node ratio at Nugent farm in 2008 and Gibbs farm in 2009.

Field	Treatment	AUHPC[a] (cm·day)	Height-to-node ratio (cm)	Dry weight (g)	AURWPC[b] (g·day)
			37DAP[C]	37 DAP	
Nugent	Gaucho	1514.88 b[d]	3.07	3.66	870.57 b
	Temik	1524.43 b	3.09	3.50	1042.81 a
	Telone	1626.77 a	3.16	3.67	1065.00 a
	P value	0.0073	0.2067	0.7569	0.0324
			35 DAP	35 DAP	
Gibbs	Gaucho	2704.71 b	2.37 b	4.06 b	648.21 b
	Temik	2872.05 b	2.67 a	4.76 b	675.09 ab
	Telone	3197.39 a	2.77 a	6.02 a	749.83 a
	P value	0.0001	0.0036	0.0008	0.0466

Cultivar effect		FM[e]	ST	FM	ST	FM	ST	FM	ST
2008	Average	1582.00	1528.72	3.15	3.07	4.08	3.14	917.24	1068.35
	P value	0.4439		0.4999		0.0308		0.0698	
2009	Average	2828.64	3020.80	2.55	2.65	4.88	5.01	718.15	663.93
	P value	0.3275		0.2795		0.7799		0.2860	

[a] Area under the height progress curve. Cotton shoot heights were measured twice at Nugent farm and three times at Gibbs farm throughout the cotton growing season.
[b] Area under the root weight progress curve. Cotton root weights were measured twice in both 2008 and 2009 throughout the cotton growing season.
[c] Days after planting.
[d] LSD(0.05) comparisons among treatments within a trial. Means in a column followed by the same letter are not significantly different.
[e] FM = FiberMax 9630B2F; ST = 4554B2RF.

Table 4.4. Effect of nematicides on cotton yield and fiber quality at Nugent farm in 2008 and Gibbs farm in 2009.

Field	Cultivar	Color grade[a]	Staple (32nds)[b]	Micronaire		Strength (g/tex)	Leaf grade	Reflectance	Yellowness	Trash (%)[c]	Length (cm)	Uniformity (%)	Lint yield (kg/ha)
													kg ha^{-1}
Nugent	Gaucho	31.13 a[d]	36.13	3.86		30.35	4.00	76.84 ab	87.25	0.38 b	2.88	81.91	967.43
	Temik	31.25 a	36.13	3.80		30.68	4.13	76.28 b	76.63	0.50 a	2.86	82.03	933.33
	Telone	27.50 b	36.13	3.78		30.88	4.00	77.33 a	89.25	0.39 ab	2.88	81.99	1007.54
	P value	0.0572	1.0000	0.3227		0.2955	0.3966	0.0072	0.4148	0.0708	0.5405	0.8991	0.2549
				FM[e]	ST								kg plot^{-1}
Gibbs	Gaucho	41.00	36.11	3.9	3.675	29.50	3.22	77.66	66.78	0.42 a	2.87	81.76	2.35 b
	Temik	41.00	36.50	3.84	3.72	29.94	3.00	77.27	66.80	0.34 b	2.88	82.02	2.56 b
	Telone	40.00	36.50	3.74	3.9	29.41	3.10	77.70	66.60	0.31 b	2.89	81.70	2.93 a
	P value	0.3759	0.1681	0.2187	0.1550	0.2599	0.2196	0.6068	0.6884	0.0321	0.3355	0.5885	0.0013

[a] Cotton fiber quality properties.
[b] Staple length (32nds of an inch).
[c] The percent of the sample surface covered by trash particles as determined by image analysis
[d] LSD(0.05) comparisons among treatments within a trial. Means in a column followed by the same letter are not significantly different.
[e] FM = FiberMax 9630B2F; ST = 4554B2RF.

Table 4.5. Effect of cotton varieties on yield and fiber quality at Nugent farm in 2008 and Gibbs farm in 2009.

Field	Cultivar	Color grade[a]	Staple (32nds)[b]	Micronaire	Strength (g/tex)	Leaf grade	Reflectance	Yellowness	Trash (%)[c]	Length (cm)	Uniformity (%)	Lint yield (kg/ha)
Nugent	FM[b]	29.33	37.08	3.81	31.35	4.00	77.96	83.92	0.41	2.95	82.02	982.95
	ST	30.58	35.17	3.82	29.92	4.08	75.67	84.83	0.43	2.79	81.93	955.92
	P value	0.3248	<.0001	0.9063	0.0050	0.3559	<.0001	0.9051	0.6748	<.0001	0.8087	0.8546
												kg plot^{-1}
Gibbs	FM	41.00	37.67	3.83	30.93	3.13	78.37	63.13	0.40	2.99	82.16	2.79
	ST	40.29	35.00	3.77	28.22	3.07	76.64	70.57	0.31	2.76	81.47	2.43
	P value	0.4275	<.0001	0.4368	<.0001	0.7517	0.0011	0.0034	0.0774	<.0001	0.0122	0.3683

[a] Cotton fiber quality properties.
[b] Staple length (32nds of an inch).
[c] The percent of the sample surface covered by trash particles as determined by image analysis
[d] FM = FiberMax 9630B2F; ST = 4554B2RF.

Table 4.6. Eigenvectors of principal components and explanatory variables for the Nugent 2008 trial.

Explanatory variable	Prin1[a]	Prin2	Prin3	Prin4
Shoot height 1st	**0.406**[b]	-0.115	0.071	-0.001
Shoot height 2nd	0.136	-0.208	**0.430**	0.058
Number of nodes	**0.377**	-0.044	-0.123	-0.119
Height-to-node ratio	**0.295**	-0.135	0.232	0.113
Fresh weight	**0.328**	-0.179	-0.172	-0.216
Dry weight	**0.374**	-0.057	-0.152	-0.188
Root weight 1st	**0.321**	-0.216	-0.059	-0.113
Root weight 2nd	-0.093	-0.002	**0.496**	0.066
Square mass	**0.323**	0.049	0.232	0.224
Root gall rating 1st	0.117	**0.514**	0.015	0.159
Root gall rating 2nd	0.096	0.029	-0.229	**0.629**
Egg counts 1st	0.155	**0.474**	0.114	-0.036
Egg counts 2nd	0.122	-0.033	**-0.386**	**0.510**
Nematode counts 1st	0.138	**0.349**	0.074	0.005
Nematode counts 2nd	0.203	**0.451**	0.064	-0.180
Nematode counts 3rd	-0.050	0.153	**-0.401**	**-0.326**

[a] Principal components.
[b] Eigenvectors with absolute values >0.30 are bold-faced.

Table 4.7. Regression analysis of cotton yield and fiber properties against principal components for the Nugent 2008 trial.

Response variable	R^2	P	Parameter estimates			
			Prin1[a]	Prin2	Prin3	Prin4
Lint yield	0.613	0.031	33.056*	-1.701	40.123*	99.106**
Color grade	0.169	0.592	0.618	0.579	-0.162	0.678
staple	0.509	0.145	0.008**	0.047	-0.257*	0.022
micronaire	0.371	0.302	-0.027	0.004	-0.003	0.060**
Strength	0.483	0.194	0.003**	0.034	-0.029	-0.124
Leaf grade	0.323	0.180	0.118*	0.013	-0.040	-0.037
Reflectance	0.546	0.021	-0.262**	-0.110	-0.327*	-0.006
Yellowness	0.261	0.689	-0.057	-1.351	5.291	2.434
Trash	0.207	0.016	0.049	-0.002	-0.016	-0.004
Length	0.514	0.042	0.024**	0.001	-0.019	-0.005
Uniformity	0.371	0.953	0.008*	-0.016	0.158*	-0.072

[a] Principal components significant at 0.05 (*), and 0.01 (**).

Table 4.8. Eigenvectors of principal components and explanatory variables for the Gibbs 2009 trial.

Explanatory variables	Prin1[a]	Prin2	Prin3	Prin4	Prin5
Shoot height 1st	**0.267**[b]	0.086	-0.029	-0.217	0.082
Shoot height 2nd	**0.279**	-0.018	0.066	0.060	0.105
Shoot height 3rd	**0.283**	-0.028	0.037	0.105	0.045
Number of nodes	0.245	-0.087	0.026	-0.155	0.007
Height-to-node ratio	0.206	0.178	-0.042	-0.202	0.150
Fresh weight	**0.275**	0.102	-0.017	-0.179	0.003
Dry weight	0.246	0.135	-0.012	**-0.280**	-0.047
Root weight 1st	0.228	0.190	-0.051	-0.207	-0.037
Root weight 2nd	0.152	-0.159	0.246	-0.073	-0.059
Nodes above white flower 1st	0.183	-0.179	0.083	0.042	0.043
Nodes above white flower 2nd	0.138	-0.200	-0.147	0.159	-0.115
Root gall rating 1st	-0.096	**-0.265**	0.241	-0.095	-0.095
Root gall rating 2nd	-0.198	-0.127	**0.287**	0.029	0.220
Egg counts 1st	-0.152	-0.036	-0.110	0.230	**-0.411**
Egg counts 2nd	-0.211	-0.056	0.060	-0.010	0.118
Nematode counts 1st	-0.073	0.051	**0.371**	0.100	**0.315**
Nematode counts 2nd	-0.087	**-0.348**	0.055	-0.174	**0.265**
Nematode counts 3rd	-0.071	**-0.343**	0.061	-0.188	**0.331**
Leaf Ca	0.180	0.041	**0.287**	0.169	-0.118
Leaf K	0.196	-0.214	-0.093	0.240	-0.023
Leaf Mg	-0.176	**0.272**	0.219	-0.136	0.018
Leaf N	0.114	-0.009	0.152	0.032	-0.129
Leaf P	-0.147	**0.319**	0.099	-0.091	0.008
Leaf S	0.129	0.041	0.099	**0.438**	0.023
Leaf Al	0.007	0.218	**-0.288**	0.215	**0.339**
Leaf B	-0.054	**0.295**	**0.272**	**0.255**	-0.008
Leaf Cu	-0.019	-0.160	**-0.399**	0.064	0.139
Leaf Fe	0.057	0.215	-0.071	0.153	**0.451**
Leaf Mn	-0.223	0.162	-0.132	-0.118	-0.045
Leaf Na	0.172	-0.028	0.018	**0.301**	0.181
Leaf Zn	-0.180	-0.009	**-0.280**	0.006	0.109

[a] Principal components.
[b] Eigenvectors with absolute values >0.25 are bold-faced.

Table 4.9. Regression analysis of cotton yield and fiber properties to principal components for the Gibbs 2009 trial

Response variable	R^2	P	Parameter estimate				
			Prin1[a]	Prin2	Prin3	Prin4	Prin5
Lint yield	0.771	0.024	11.498**	-0.087*	0.004	-0.040	0.177**
Color grade	0.161	0.663	-0.188	-0.098	-0.128	0.169	0.293
staple	0.298	0.259	-0.033	-0.080	-0.154	-0.395	0.372
micronaire	0.295	0.267	0.005	-0.009	-0.036	-0.014	0.060
Strength	0.242	0.405	-0.125	-0.118	-0.208	-0.203	0.293
Leaf grade	0.193	0.556	0.012	-0.060	-0.036	-0.022	-0.008
Reflectance	0.417	0.078	0.055	-0.101	0.210	-0.416*	0.167
Yellowness	0.507	0.023	0.730*	-0.293	-0.166	1.667*	-0.833
Trash	0.406	0.088	-0.005	-0.025**	0.013	0.004	0.004
Length	0.367	0.135	-0.004	-0.010	-0.013	-0.037*	0.032
Uniformity	0.275	0.313	-0.018	-0.064	-0.020	-0.225*	0.069

[a] Principal components significant at 0.05 (*), and 0.01 (**).

Table 4.10. Effect of different types of nematicides and seed treatment on cotton lint price and adjusted revenue at Nugent farm in 2008 and Gibbs farm in 2009.

Treatment	Lint Price				Adjusted revenue			
	Nugent 08		Gibbs 09		Nugent 08		Gibbs 09	
	FM[b]	ST	FM	ST	FM	ST	FM	ST
Gaucho	0.67	0.66	0.66	0.66 b[a]	487.1 a	444.2	472.7	356.8
Temik	0.67	0.65	0.67	0.67 a	468.3 a	400.0	466.7	400.5
Telone	0.68	0.66	0.67	0.67 a	382.6 b	414.6	496.5	460.4
P value	0.1517	0.4059	0.7083	0.0194	0.0051	0.6335	0.6720	0.0615

[a] LSD(0.10) comparisons among treatments within a trial. Means in a column followed by the same letter are not significantly different.
[b] FM = FiberMax 9630B2F; ST = 4554B2RF.

CHAPTER 5

SUMMARY AND CONCLUSIONS

Meloidogyne incognita is a major parasite on cotton. The management of *M. incognita* is a crucial part of cotton production. With appropriate nematode management strategies, cotton yield can be improved. However, very little data has documented the effect of *M. incognita* on fiber quality. Fiber quality has become an increasingly important issue as it affects the value of cotton lint. Some mills only accept cotton with specific standards of fiber quality. Therefore, fiber quality is extremely important to cotton economics, in addition to cotton yield.

In greenhouse trials, the effects of *M. incognita* on key physiological processes along with cotton growth characters were examined. Three nematode infection levels were created by infesting soil planted to cotton with different numbers of eggs of *M. incognita*. Two commercial cotton varieties and two cotton breeding lines were used in greenhouse trials to compare their responses to *M. incognita* infection.

The results from greenhouse trials showed that with infection by *M. incognita*, cotton vegetative growth, in terms of height-to-node ratio, shoot dry weight, leaf expansion; as well as cotton reproductive growth, in terms of number of bolls and boll weight, were all significantly reduced. This may have resulted from reduced photosynthetic products, reduced root uptake and delayed plant development. The reduction in photosynthetic rate caused by *M. incognita* infection was usually accompanied by reduced transpiration rate, reduced stomatal conductance, as well as increased leaf temperature and sub-stomatal CO_2 concentration, which indicated that

the damage caused by *M. incognita* resembles drought stress. These results indicated that fiber development, which requires sufficient organic and inorganic nutrients, may be interrupted by *M. incognita* infection. Of the varieties and breeding lines tested, 120B1R1 showed a very promising resistance to *M. incognita*, especially in terms of suppressing nematode reproduction in root tissues.

Field experiments were conducted at five different field sites over two growing seasons. Cotton yield and fiber quality, as well as plant growth, root damage and nematode populations were assessed. Different-sized populations of *M. incognita* were created by applying different nematicides at different rates to respective plots. In addition to the nematicide treatments, site-specific management zones, with proven effect on cotton yield, were also used at three out of the five field sites. At these three fields with management zones, two rates of Temik and two rates of Telone were applied in both zones. Seed treatments AVICTA Complete Cotton and AERIS Seed Applied System + Trilex were applied only at Perryman farm.

Results from these field trials showed that with different nematicide types, rather than different nematicide rates, significantly different levels of nematode population, nematode infection, and reproduction were created successfully. The greatest nematode population occurred with seed treatments, and the lowest nematode population was achieved with Telone treatments. The difference between low and high rates of nematicides was not significant. As a result, plant vegetative growth was significantly improved in plots fumigated with Telone. Plots treated with Temik resulted in better plant growth than seed treatments, but the results were not as good as in plots where Telone was applied.

Cotton yield was improved where Telone was applied as compared with other treatments in most cases. However, in 2009 at two Windhausen farms, cotton yield was reduced by Telone

treatments, especially the high rate of Telone. Since the damage in cotton caused by *M. incognita* was shown to resemble drought stress in greenhouse trials, with the excessive rainfall in 2009, plant growing conditions were improved; therefore less damage was caused by *M. incognita*. Also, cotton yield might be less sensitive to *M. incognita* infection than plant growth. Therefore, cotton yield was not affected, while plant growth was. Cotton fiber quality was not consistently or significantly affected by different treatments in these field trials. A few fiber properties were affected in some trials, but they did not differ according to nematode-associated disease severity levels. Therefore, they may have been affected by other factors rather than root-knot nematodes. These effects were also not consistent in other trials. Therefore, cotton fiber quality may be less sensitive to *M. incognita* infection than yield.

The management zone effect was usually not significant at these three field sites. The most obvious zone difference was seen in the Perryman trials. In these trials, plant growth and nematode population were both significantly higher in the high-risk zone. This indicated that the zone difference may be due to some environmental factors other than nematodes, because *M. incognita* has been shown in both greenhouse and field trials to reduce plant growth. Cotton growth, yield, and fiber quality all responded the same to nematicide in different management zones.

At both the Nugent and the Gibbs farm, two varieties of cotton were planted to plots treated with three chemical treatments, including Gaucho Grande seed treatment, Temik 15G and Telone II. In these trials, plots fumigated with Telone had significantly improved plant growth, but only increased yield in the Gibbs 2009 trial. This may have occurred because at the Nugent farm there was generally a lower nematode population than in other fields. Also, irrigated conditions may have provided the cotton with an optimum growing environment. plus cotton

yield seems less affected by *M. incognita* than cotton growth. In the Gibbs 2009 trial, the most significant difference among treatments was shown in plant growth, nematode population, root damage, and nematode reproduction than any other trial. Yield was significantly improved by Telone in both fields. However, none of these trials showed an effect of treatments on fiber quality. The reason fiber quality was not affected by *M. incognita* infection might be because the developing bolls are nutrient sinks and compete with *M. incognita* for nutrient transport. If developing bolls are stronger nutrient sinks, or if *M. incognita* are stronger nutrient sinks but sufficient nutrients exist, fiber development and quality may not be affected.

There was a significant difference in fiber quality between the two cotton varieties used at the Nugent and Gibbs farms. Many of the fiber properties were significantly higher in lint from the FiberMax variety, which was considered the susceptible variety in this study. Although not significant, plots planted to the FiberMax variety had a higher nematode population and disease severity than plots planted to Stoneville seed in both trials. Since none of the other trials showed an increased fiber quality with higher level of *M. incognita* infection, this difference in fiber quality between varieties may only be due to their genetic differences.

The principal component analysis was conducted to analyze the direct relationship of response variables (yield and fiber properties) with all the explanatory variables (plant growth, nematode population, soil nutrients, etc.). The results showed that the effect of *M. incognita* on fiber quality was not significant in most cases and also not consistent in different years. It was not clear whether the effect of nematode on overall fiber quality was positive or negative.

The values of fiber properties changed over a relatively small range. However, with a small change, the value of the fiber could drop under the USDA standard and cause an economic penalty to the growers. Therefore, lint price based on market average and fiber quality, and

adjusted revenue based on expenditure, yield, and fiber quality were analyzed for all trials. The lint price from all five fields and two growing seasons showed no significant difference with different nematicide treatments. The average fiber price for the 2 years was $0.65/lb. The overall fiber price for Perryman and Windhausen farms was below the average, and the price for Nugent and Gibbs farms was above the average. None of the treatments changed this situation on a numerical level. Adjusted revenue was significantly lower with Telone treatments in fields with lower nematode pressure or better environmental conditions, e.g., higher soil moisture. Therefore, Telone application may be more profitable in fields with high nematode pressure and more environmental stress.