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Performance of upland cotton (Gossypium hirusutum) in reniform

(Rotylenchulus reniformis) nematode infested soils as

affected by variety and seed treatment

By

Harry Randall Smith

A Dissertation Submitted to the Faculty of Mississippi State University in Partial Fulfillment of the Requirements for the Degree of Doctor of Philosophy in Agronomy in the Department of Plant and Soil Sciences

Mississippi State, Mississippi

May 2016

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Harry Randall Smith

Performance of upland cotton (Gossypium hirusutum) in reniform

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affected by variety and seed treatment

By

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Title of Study:Performance of upland cotton (Gossypium hirusutum) in reniform<br/>(Rotylenchulus reniformis) nematode infested soils as affected by<br/>variety and seed treatment

Pages in Study: 171

Candidate for Doctor of Philosophy

Reniform nematode (*Rotylenchulus reniformis* Linford and Oliveira) currently infests about 36% of the Mississippi *G. hirsutum* acres causing economic losses of \$130 million annually. For more than 40 years nematodes, including *R. reniformis*, have been managed using an at-planting treatment of Temik 15G or with soil fumigants like Telone II. With the label loss of Temik 15G and expense of soil fumigants, there is a need to develop an integrated nematode management program centered around nematicide seed treatments (NST) with and without foliar applications of Vydate C-LV. In addition there is a need to better understand how new cotton cultivars provide improved growth, development and yield in nematode infested fields. Results from research at Auburn and Mississippi State Universities revealed tested varieties responded positively to NST and improved growth and yield without NST was variety specific especially early in *G. hirsutum* development (between nodes 1-9). Cutivars Phy 499, FM 1740 and Stv 5458 showed the greatest nematode tolerance while Phy 375 WRF had the least tolerance, benefitting greatly from NST. Trials involving NST with and without Vydate C-LV indicated yield of plants treated with Temik 15G was greater than plants treated with NST treatments. Aeris + Votivo with and without Vydate C-LV provided better plant growth and yield than Aeris alone or with Vydate C-LV. Relative to yield Vydate C-LV treatments increased pounds of lint cotton/acre across all treatments. There were no differences in fruit retention at fruiting site one during the square period with fruit loss primarily occurring between bloom and open boll. Vydate C-LV treatments increased overall fruit retention compared to all nematicide seed treatments making them comparable to Temik 15G.

#### DEDICATION

I dedicate this dissertation to my father, Alfred Randolph Smith Jr, who always provided me with a can do attitude and perseverance despite the obstacles and began my road in production agriculture first as a County Agent in Claiborne County and then as a producer and owner of one of the largest retail/wholesale pecan (Smith's Pecan) operations in the country. I further dedicate this work to family members who are no longer with us but who made significant contributions to my life and Mississippi agriculture. First my mother, Mrs. JoAnn Smith, my maternal grandparents (Mr. and Mrs. Harry Thomas Shuff) and my paternal grandparents (Mr. and Mrs. Alfred Randolph Smith) and my paternal aunt (Mrs. Elizabeth Rushing). These people began my knowledge and appreciation of plant science, whether horticulture, forages or row crops, at a very early age. This knowledge has enabled me to have a great understanding of plant developmental stages that has allowed me to address crop changes relative to specific conditions quickly to produce a successful crop. They further provided me with an understanding of fairness, conviction and honesty.

#### ACKNOWLEDGEMENTS

First, I thank Dr. Richard Harkess, my major advisor, for his support and direction in my quest to complete this task. Dr. Harkess is a person whom I greatly respect both as a professor and person. When things would get tough, Dr. Harkess would always provide an attitude adjustment that seemed to be tailored for my personality to make me determined to complete the task. I not only gained knowledge in crop physiology but also a philosophy of being a good person. I express my deepest thanks to Dr. Harkess.

Second, I am greatly indebted to my co-major advisor, Dr. Gary Lawrence, who as a nematologist has instilled an even greater interest in nematology and the integrated crop management that surrounds this animal.

Third, I am extremely thankful for the assistance and direction provided by Dr. Patricia Knight and Dr. Lane Porter.

Fourth, I am very thankful to Mrs. Carolyn Conger who made sure I did not quit when things got difficult. She also greatly assisted me in the field while collecting massive amounts of data and has been very supportive while analyzing and summarizing the data.

Fifth, I thank the entire nematology team at Mississippi State University who aided in data collection and plant mapping. Their selfless attitude and dedication to the team did not go unnoticed.

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Last, I thank my committee members, Mississippi State Department of Plant and Soil Sciences, Mississippi State University Extension Service and Auburn University who made all of this possible.

Special thanks to Bayer Crop Science and Phytogen Seed for sponsoring the project.

Other people who have greatly shaped my philosophies include Mrs. Janice Jones, Mr. James Wiley Nelms, Dr. William H. McCarty, Dr. Allen M. Blaine, Dr. Gordon Andrews, Dr. Billy Moore, Dr. Donnie Miller, Dr. Roger Leonard and Dr. Johnny Jenkins from the University sector. Those from corporate sector include, Dr. Tom Kerby, Mr. Richard Shaw, Mr. Steve Lee, Mr. Rick Turnage, Mr. Calvin Bowlin, Dr. Brewer Blessit, Dr. Dan Poston, Mr. Heath Hughes, Mr. Mike McCormick and Mr. Jim Braucht. From the grower sector special thanks to Mr. Bernie Jordan, Mr. Mike and Ben Lamensdorf, Milton Parish and all the growers in the southwest and southeast Mississippi who have supported me in this process.

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#### CHAPTER I

#### INTRODUCTION

Cotton (*Gossypium hirsutum*) is the world's most important natural fiber crop. The United States *G. hirsutum* lint (fiber) production accounts for one-quarter of world supply (USDA-Agricultural Marketing Service, 2007). In Mississippi, *G. hirsutum* remains a significant agronomic crop accounting for 1.1 million hectares in 2013 (Mississippi Agricultural Statistical Service, 2013). Since 1990, the *G. hirsutum* industry has undergone several positive changes including boll weevil eradication, introduction of genetically modified *G. hirsutum* cultivars and development of more efficient harvest and planting equipment (USDA-Agricultural Marketing Service, 2007). These milestones lowered inputs facilitating management of other important problems like plant parasitic nematodes.

The predominant plant parasitic nematode that has become the most damaging pathogen to *G. hirsutum* is the reniform nematode (*Rotylenchulus reniformis* Linford and Oliveira). *Rotylenchulus reniformis*, first described in 1931 (Linford and Oliveira, 1940), has become a widely distributed tropical and subtropical pest throughout the United States *G. hirsutum* producing region (Heald and Robinson, 1990; Kinloch and Sprenkel, 1994; Lawrence and McLean, 1996 ab; Star, 1998; and Koenning, et al., 1999). *Rotylenchulus reniformis* depends on successful formation of feeding sites in *G. hirsutum* roots that serve as site of nourishment. It has been well documented the vermiform female of *R. reniformis* penetrates *G. hirsutum* roots indiscriminately until 50% of its anterior body enters the root making it a semi-endoparasite. It establishes feeding sites near the root pericyle where it creates synecia from altered pericycle cells (Jones and Dropkin, 1975). Because of this feeding mechanism, *R. reniformis* cause uniform stunt across a field, making it difficult to visually identify. In limited regions, they cause interveinal chlorosis and yield loss (Lawrence and McLean, 2001). *Rotylenchulus reniformis* is also known to affect *G. hirsutum* by reducing yield, boll size and lint percent (Cook et al., 1997b; Jones et al., 1959). It has been further shown, *G. hirsutum* plants respond poorly to normal agronomic management practices i.e. irrigation and fertilization (Birchfield and Jones, 1961). In addition to direct impacts of feeding, *R. reniformis* provide portals for introduction of several soil-borne pathogens including *Fusarium oxysporum* f. sp. *vasinfectum, F. solani, Rhizoctonia solani* and *Thielaviopsis basicola* (Palmateer et al., 2004).

Since 1960, *R. reniformis* began manifesting adaptive ability to survive colder environments allowing movement through much of the eastern half of the *G. hirsutum* producing region (Heald and Robinson, 1990) and as far north as Lubbock, Texas, and Missouri bootheel (Heald and Thames, 1982; Wrather et al., 1992). Today, *R. reniformis* has been identified and associated with *G. hirsutum* yield loss in Mississippi, Alabama, Tennessee, Texas, Missouri, Florida, North Carolina, Louisiana, South Carolina, Arkansas and Georgia (Koenning. et al., 1999), accounting for 7% annual yield loss and nearly \$126 million loss to the *G. hirsutum* industry in 2008 (Blasingame et al., 2009), and 11.7% in 2014 (Lawrence et al., 2015), resulting in approximately \$70.0 million in economic losses. In Mississippi alone, *R. reniformis* was responsible for annual losses of 235,398, 252,023, 56,378 and 58,000 bales of G. hirsutum in 2004, 2005, 2011 and 2014, respectively (Blasingame and Patel, 2004; 2005; 2011; Lawrence et al., 2015). Lawrence, et al. (2002) reported more than 32% of cotton acres in Mississippi were infested with R. reniformis increasing the threat to G. hirsutum yields (Lawrence and McLean, 1995 a and b). Gazaway and McLean (2003) further reported R. reniformis infested more than 36% of the Alabama G. hirsutum production area and is increasing. Diez et al. (2003) reported that a population shift began in 1986 from root knot nematode (*Meloidogyne incognita*) toward *R. reniformis* infestation and was accomplished by 2004. A primary reason for population shift was due to ability of *R. reniformis* to reduce *M*. incognita egg hatching, thereby reducing secondary generation infection (Diez et al., 2003). The characteristics promoting rapid spread is ability of *R. reniformis* to reproduce in a broader range of soil types than *M. incognita* (Koenning et al., 1996; Widmer et al., 2002; Gazaway and McLean, 2003; Moore and Lawrence, 2013). It also has ability to survive and promote yield loss under drought conditions (Herring et al., 2010), survive long periods in fallow fields by tolerating dehydration of its egg masses followed by rehydration under favorable conditions (Heald and Thames, 1982; Koenning et al., 1996) and can spread completely across a field in one season due to fecundity and ability to move by equipment and irrigation (Moore et al., 2010; Monfort et al., 2008). R. reniformis also has ability to survive deep in the soil profile (Moore et al., 2010; Lee et al., 2003; Robinson, 2005 a and b; 2005b; Heald and Thames, 1980). Moore et al. (2010) reported finding these parasites at a depth of 91 cm and moving horizontally a distance of 200 cm in one season. This was further verified by Lee et al. (2015) where it was reported that R. reniformis was found 120 cm deep. Heald and Thames (1980) reported

finding *R. reniformis* at depths of 1.75 m from soil surface and these populations correlated to the *G. hirsutum* root zone. However, it has been reported *R. reniformis* occurred at deeper soil depths despite presence of *G. hirsutum* roots (Lee et al., 2015; Robinson et al., 2005a). Lee et al. (2003) further reported *R. reniformis* could fluctuate to depths of 1.2 m throughout the season depending on environmental conditions. Newman and Stebbins (2002) and Robinson et al. (2005b) found these deep nematode populations could reduce *G. hirsutum* yields, but yields could be increased using nematicides like Temik 15G applied as a side-dress application.

In addition to surviving well at deeper soil depths, *R. reniformis* it can survive in a wide array of soil textures. Starr et al. (1993) reported only 12% of samples possessing *R. reniformis* had a sand content greater than 40%. Robinson et al. (1997) further reported *R. reniformis* at higher incidence levels in soils with textures of higher silt and clay. However, Gazaway and McLean (2003) first reported a greater presence of *R. reniformis* occurring in coarser textured soils. Further attributes of *R. reniformis* survival is ability to rebound quickly following rotation to corn (*Zea mays*). Davis et al. (2003) and Windham and Lawrence (1992) reported that following a one year corn rotation with *G. hirsutum* resulted in higher *R. reniformis* population than where rotation was not followed by bloom of *G. hirsutum* growth. This was further verified by Lee et al. (2015). Further facilitating increased populations of *R. reniformis* is its wide host range of 314 plant species surveyed to date (Robinson et al., 1997).

Management of *R. reniformis* in *G. hirsutum* was primarily with Temik 15 G (a main-stay nematicide/insecticide for over 40 years). However, with removal of label use of Temik 15G in 2012, it became evident and necessary to evaluate other means to

produce cotton in *R. reniformis* infested soils. An integrated approach to improve efficacy of nematicide seed treatments (NST) is needed. This integrated crop management approach involves better understanding of how to improve overall G. hirsutum health while growing in R. reniformis infested soils. Integrated crop management is important because crop rotation is not viable in all G. hirsutum producing areas (Davis et al., 2003), resistant cultivars are not commercially available, little information is available on tolerance in commercial varieties (Koenning et al., 2000; Starr et al., 2007) and nematicide applications are expensive with environmental concerns. Attempts have been made to reduce need for and improve efficacy of nematicides in R. *reniformis* infested soils primarily with GIS/GPS systems as a part of integrated G. hirsutum management programs (Herring et al., 2010; Overstreet et al., 2010; Greer et al., 2009; Lawrence et. al, 2008; Wolcott et al., 2008; Monfort et al., 2007; Ellis et al., 2005; Wolcott et al., 2005). In addition, work has been conducted in an integrated G. hirsutum management program to showcase yield improvement of G. hirsutum grown in different soil types infested with R. reniformis relative to irrigation. Herring et al. (2010) reported coarser texured soils infested with R. reniformis produced lower G. hirsutum yields than soils possessing finer textures, but the lower yielding soil textures could be improved via timely irrigations. They further demonstrated effects of *R. reniformis* on cotton yield were independent of irrigation, but dependent on soil texture. Similar results were reported by Moore and Lawrence (2013) and Davis et al. (2014). Widmer et al. (2002) further demonstrated increased soil organic matter improved overall plant health and performance in nematode infested soils by changing soil microflora which can reduce parasitic nematode populations. Therefore, understating different parameters of G.

*hirsutum* management as it improves *G. hirsutum* plant health can improve performance in *R. renifo*rmis infested soils.

Further knowledge of commercial G. hirsutum germplasm performance in R. reniformis infested soils is pertinent in developing a successful integrated G. hirsutum management program. Numerous studies have been conducted to evaluate performance of commercial G. hirsutum varieties in nematode infested soils as related to tolerance. Tolerance can be defined as the plant's ability to sustain itself in nematode presence without dying or having serious injury or yield loss (Agrios, 1978). Tolerant plants support nematode reproduction while displaying acceptable yields compared to susceptible plants (Koenning et al., 2000). Cook et al. (1997 a) stated G. hirsutum tolerance might be a management possibility but little information exists relative to R. *reniformis*. Since 1988, eleven *M. incognita* tolerant breeding lines have been released (Jones et al, 1988; Cook et al., 1997a; Cook et al., 1997b; Cook and Robinson, 2005) to M. incognita. These varieties yield well in the M. incognita infested fields of their production regions. However, according to Koening et al. (2001), these varieties might not be adapted to a wide geographic area. Wheeler, et al. (2014) reported a positive economic interaction between nematicides plus foliar applications of Vydate C-LV<sup>®</sup> and variety in *M. incognita* populations. Usery et al. (2004; 2005), Legee et al. (2007) and Blessit et al. (2012) reported several varieties showed tolerance in high R. reniformis infested soils. Earlier maturing varieties showed greater tolerance to R. reniformis providing higher yields and lower nematode feeding activity in the roots (Usery et al., 2005). However, Blessitt et al. (2012) demonstrated no relation between maturity and G. *hirsutum* performance in *R. reniformis* infested soils but stated that six of thirteen *G.* 

hirsutum varieties tested showed tolerance. Further work evaluating commercial variety performance in nematode infested soils was reported by Phipps and Eisenback (2005) and Davis (2005) as it related to *M. incognita*. This group further showed no difference to nematode species infestation related to maturity in G. hirsutum varieties. Koenning et al. (2005), however, reported late maturing varieties performed better than early maturing varieties in soils infested with the Columbia lance nematode (*Hoploaimus columbus*) while Williams et al. (2004) reported similar findings to *M. incognita*. Phipps and Eisenback (2005) further reported net dollar return was greater when using tolerant G. *hirsutum* varieties planted in *M. incognita* infested fields. They also reported nematicides were still economically beneficial when used with tolerant varieties. There are several public sector varieties that show promise in highly infested nematode soils (Davis et al., 2010). The only advanced technology (Widestrike/Roundup Flex or BG 2/RF) containing cotton variety evaluated that has shown nematode tolerance is Phy 367 WRF. McPherson and Rush (2011) cited that Phy 367 WRF showed excellent response in M. incognita infested soils despite not being treated with nematicides.

Nematicides continue to be an important segment of an integrated *G. hirsutum* management program that allows cotton to be successfully produced in nematode infested soils. Since 2003, the *G. hirsutum* industry began moving away from granular, at-planting treatment with Temik 15G for nematode control. Nematode seed treatments today have replaced Temik 15G in the industry. Padgett and Overstreet (2004) reported some NSTs were as effective as Temik 15G when compared at nematicide rate of 0.75 lbs ai/Ac and some seed treatments reduced galling over untreated check but did not improve maturity or yield. This indicates lack of longevity of seed treatments compared

to Temik 15G and necessitates need for additional management options to improve cotton performance in nematode infested soils. Kirkpatrick and Monfort (2004) reported NST did not differ in nematode management from Temik 15G from 14 to 35 DAP. In addition, they reported NST applied at 100 g (a.i)/kg of seed was similar to Temik 15G applied at 0.75 lbs ai per Ac. Monfort et al. (2004) reported root knot nematode numbers and gall numbers were reduced using NSTs similar to using Temik 15G. A major concern of NST was lack of early season insect control compared to Temik 15G. Brown et al. (2008) reported tobacco thrip (*Frankliniella fusca*) damage occurring in early developmental stage of *G. hirsutum* growing in nematode soils reduced early root growth and yield. However, the group did not evaluate loss of maturity as a result of combined effects from nematodes and thrips. This research led the industry to combine seed treatments containing insecticides with nematicides or mandating an over-top application for insect management.

To further enhance and improve *R. reniformis* management of NST treatments beyond 35 days after planting, foliar applied Vydate C-LV<sup>®</sup> has been shown to be an excellent tool used in conjunction with older nematicide products (Lawrence and McLean, 2000; 2002; 2003). Vydate C-LV<sup>®</sup> with nematicide/insecticide properties remains a viable tool in managing *G. hirsutum* nematodes because of ease of foliar application and phloem transmission to root system (Hsu and Kleier, 1996). This tool becomes crucial since *R. reniformis* obtains maximum population densities when *G. hirsutum* is in its peak reproductive phase (Lawrence and McLean, 1995 a and b; 1996 a and b; 1997). The additional plant stress from parasitism by *R. reniformis* can result in

reduced yields and requires additional treatments beyond those obtained by NST treatments.

Understanding *G. hirusutum* growth and development is critical in understanding biotic and abiotic stress effects while implementing management strategies for maximizing yields and profits. *Gossypium hirusutum* possesses a unique fruiting pattern of simultaneous reproductive and vegetative growth which makes *G. hirsutum* much different in growth pattern compared to other row crops. This growth mechanism makes *G. hirusutum* an ideal plant in which to evaluate and quantify stresses due to nematodes (Jenkins and McCarty, 1995; Kerby et al., 1987; Smith et al., 1996; Smith and Turnage, 1998).

Gutherie and Kerby (1993) reported *G. hirusutum* growth maintains a record of its response to environmental conditions and management inputs which can be traced by observing its vegetative structure and fruit distribution. The vegetative and reproductive growth distribution can be quantified by plant mapping processes. Biotic or abiotic stresses can be placed on a developmental time-line by denoting where the symptoms occurred on the plant. Early-season conditions are recorded in vegetative growth and square retention levels while mid-season effects are observed in internode lengths and boll retention. Late-season influences impact location of last harvestable boll and degree of secondary growth. Plant mapping importance has been well documented (Jenkins and McCarty, 1995; McCarty et al., 1994, Albers, 1993; Hake et al., 1990). In-season plant mapping has been used extensively to quantify treatment effects in *G. hirusutum*. Smith and McCarty (1996) used in-season plant mapping to demonstrate Temik 15G effectiveness applied at-planting and as a side-dress in *G. hirusutum* growing in *R*.

*reniformis* infested soils. From this methodology, Smith and McCarty (1996) were able to capture fruiting pattern differences, growth differences, maturity and yield resulting from the treatments. Turnage and Smith (1998) further used in-season plant mapping to demonstrate how Temik 15G performed compared to Acephate 15G under heavy thrips pressure of PM 1215 *G. hirusutum* variety based on fruit retention, height to node ratios, earliness and yield grown in *R. reniformis* infested soils. Lawrence et al. (1998; 2001; 2002) and Lawrence and McLean (2002) further showed influence of nematicide treatments on *G. hirusutum* in *R. reniformis* infested soils via plant mapping processes.

The objectives of these studies were to evaluate *G. hirsutum* growth and development using plant mapping processes comparing NST to Temik 15 G and foliar applications of VydateC-LV<sup>®</sup> (oxamyl) in combination with the NST in *R. reniformis* infested soils; to evaluate performance of five commercially available *G. hirsutum* varieties with and without NST to determine if varieties are tolerant to *R. reniformis*; and to determine treatment efficacy population thresholds of *R. reniformis* using controlled environments to verify field findings.

#### CHAPTER II

#### LITERATURE REVIEW

# Integrated management of cotton grown in reniform nematode infested soils as affected by nematicides and varieties

#### **General Introduction**

Presently, suppression of *R. reniformis* in cotton is with use of nematicides (granular, fumigant, biological, seed treatments and foliar). The granular product, Temik 15G (aldicarb), a mainstay in the cotton industry for over 40 years and recognized for its superior at-planting and side-dress insecticide and nematicide properties, was removed from market by Bayer Crop Science. The decision was preceded by launch of several nematicide seed treatments (NSTs) that are environmentally friendly with more favor with the Environmental Protection Agency. Lawrence et al. (1990) and Lawrence and McLean (2001), from an aggressive testing program, saw positive economic returns when using fumigant nematicides in heavily infested *R. reniformis* fields; however, this practice is costly, application takes considerable time and requires special equipment. In lieu of this, a move in 2003, to treat cotton seed with chemicals and/or biologicals with known nematicide properties occurred.

#### Nematicide Seed Treatments

Since 2003, the cotton industry has been moving away from granular, at-planting treatment of Temik 15G for nematode control. Nematicide seed treatments today have

replaced Temik 15G. Padgett and Overstreet (2004) reported some NSTs were as effective as Temik 15G when compared at its nematicide rate of 0.75 lbs ai/Ac and some seed treatments reduced galling over untreated check, but did not improve maturity or yield. This indicates lack of longevity of seed treatments compared to Temik 15G and necessitates need for additional management options to improve G. hirsutum performance in nematode infested soils. Kirkpatrick and Monfort (2004) reported NSTs did not differ from Temik 15G from 14 to 35 DAP. In addition, they reported NSTs applied at 100 g (a.i)/kg of seed was similar to Temik 15G at 0.75 lbs ai/Ac. Monfort et al. (2004) reported root knot nematode and gall numbers were reduced using NSTs similar to Temik 15G. A major concern of NST was a lack of early season insect control obtained from Temik 15G. Brown et al. (2008) reported tobacco thrip (Frankliniella *fusca*) damage occurring in early cotton developmental stages combined with nematode infestation reduced early root growth and yield. However, the group did not evaluate harvest maturity delays as result of combined nematode and thrips effects. This research led the industry to combine seed treatments containing insecticides with nematicides or mandating over-top applications for insect management.

To further enhance and improve *R. reniformis* management of NST treatments beyond 35 days after planting, foliar applied Vydate C-LV<sup>®</sup> has been shown to enhance nematode management combined with older nematicide products (Lawrence and McLean, 2000; 2002; 2003). Vydate C-LV<sup>®</sup> with nematicide/insecticide properties remains a viable tool used in managing nematodes in *G. hirsutum* because of ease of foliar application and phloem transmission to root system (Hsu and Kleier, 1996). This tool becomes crucial since *R. reniformis* obtains maximum population densities when cotton is in peak reproductive phase (Lawrence and McLean, 1995 a and b; 1996 a and b; 1997). The additional plant stress from parasitism by *R. reniformis* can result in reduced yields and requires additional treatments beyond what is obtained by NST treatments.

Today, the primary seed treatments for nematode suppression in cotton include; Aeris<sup>®</sup> (thiodicarb), Avicta<sup>™</sup> (abamectin), Votivo<sup>®</sup> (*Bacillus firmus*) and N-Hibit<sup>®</sup> (*Erwinia amylovora*) (Woodard et al., 2008; Kirkpatrick et al., 2011; Overstreet and Kirkpatrick, 2011). Other companies are emerging with experimental biological products for nematode suppression in crops that will also be used as a seed treatment.

Aeris<sup>®</sup> Seed Applied System (imidachloprid + thiodicarb @ 0.375 mg (ai)/Lb of seed)

This product is listed for suppression of root knot and reniform nematodes as well as *Frankliniella fusca* (tobacco thrips), *Sericothrips variabilis* (soybean thrips), *Thrips tabaci* (onion thrips) and *Aphis gossypii* (cotton aphids). It provides an additional option of having Trilex<sup>®</sup> (seed treatment fungicide) for control of *Rhizoctonia, Pythium, Thielaviopsis* and *Fusarium*. Graham et al. (2007) reported AERIS<sup>®</sup> Seed Applied System increased plant height, leaf area, white-bloom count and yield when compared to Avicta<sup>™</sup> which was also reported by Kemerait et al. (2006; 2007; 2008). However, Kemerait et al. (2007; 2008) reported Temik 15G provided better yields and return on investment (ROI) when compared to seed treatments in nematode infested soils.

Avicta Complete Cotton<sup>TM</sup> (abamectin + cruiser + dynasty)

The abamectin portion of Avicta<sup>™</sup> is a macrocyclic lactone produced by *Streptomyces avermitilis* (Faske and Starr, 2007). Monfort et al. (2004), Kirkpatrick and Monfort (2004) and Phipps and Eisenback (2007) reported Avicta<sup>™</sup> provided similar nematode control as Temik 15G. This was disputed by Kemerait et al. (2006; 2007; 2008) who found Avicta<sup>™</sup> performed at a lower level than AERIS<sup>®</sup> or Temik 15G and provided a negative ROI.

#### Votivo<sup>®</sup> (Bacillus firmus)

VOTIVO<sup>®</sup> is a biological nematicide believed to protect roots from early season nematode damage by colonizing roots and immediate root environment promoting plant growth which characterizes this product as a Plant Growth Promoting Rhizobacteria (PGPR). Five to ten million spores are applied per seed and once the bacteria begins to grow (activated by temperature and water) expands exponentially with root growth as the bacterium uses root exudates as a food source. It is believed bacterium colonization of root receptor sites used by nematodes reduces root finding by nematodes (Riggs and Bugg, 2011; Bugg, 2010).

### N-Hibit and Messenger<sup>®</sup> (Erwina amylovora)

Both products are classified as harpin proteins which activate natural stressdefense mechanisms improving plant vigor and health. Harpin proteins were first isolated from *Erwinia amylovora* (Wei et al., 1992) and shown to promote gene expression involved in hypersensitive response, plant growth, stamina, increased yields, improve shelf-life and induce systemic plant defense (Wei and Berr, 1996). N-Hibit<sup>®</sup> is seed applied while Messenger<sup>®</sup> is applied foliarly. Kirkpatrick et al. (2005) working with *M. incognita* did not show differences in plant growth, but did show reduction of galls and reduced reproduction. Kirkpatrick et al. (2005), in growth chamber, reported increased plant height, plant biomass, and node number from harpin proteins applied to seed or foliage. French et al. (2006 a and b) showed positive yield increases comparable to Temik 15G and Avicta.

#### Historical reasons for loss of aldicarb (Temik 15G) manufacture in the U.S.

Aldicarb (2-methyl-2-(methylthio)-propionaldehyde O-methylcarbamoyloxime) had been used on cotton since 1970 in the United States for control of sucking, piercing insects and plant parasitic nematodes. Temik 15G is highly toxic with an oral LD<sub>50</sub> of 0.3-0.9 mg/kg (Cox, 1992). When misused it can cause death to mammals (Center for Disease Control, 1986; 1999) and continue mortality within food chain due to tissue persistence. Balcomb et al. (1982) observed high mortality levels from Temik 15G in sparrow and blackbird populations of 80 and 40% respectively. In 1986, California watermelons illegally treated with Temik 15G during 1985 promoted one of the largest poisonings in North American history where nearly 2,000 people became ill (Green et al, 1987; Goldman, 1990). In this incident there was one fatality and several pregnant women gave birth to stillborn babies. This was followed by illnesses in Louisiana during 1998 when Temik 15G placed in a pepper container was used to season a salad resulting in 20 illnesses (Center for Diseased Control, 1999). Aldicarb related posionings in Nebraska and British Columbia have been cited to occur from illegal applications in cucumbers (World Health Organization, 1991). In 1991, above tolerance levels were found in bananas prompting the removal of bananas from the Temik 15G label. In 1979, Temik 15G residues were detected in well water in the New York's potato-growing region after only four years of use. By 1986, 2,500 wells were found to be contaminated with unacceptable levels of Temik 15G which prompted a cease use (Jones and Marquardt, 1987). This led the Environmental Protection Agency (EPA) under The Safe

Drinking Water Act to develop the Maximum Contaminant Level Goal (MCLG) which is the contamination level where no known detriment occurs for man (U.S. E. P. A., 1998). The level was established at 1.0 part per billion, especially for Temik 15G. Following these reports, 26 states, including Mississippi, were cited for Temik 15G well water contamination (U.S. E. P. A., 1991). From these studies, solubility of Temik 15G and its metabolites and contaminants (aldicarb sulfoxide, aldicarb sulfone, aldicarb oxime, dichloromethane and N-nitrosaldicarb) were documented (U.S. E. P. A., 1988). Pacenka et al. (1987) further reported enormous solubility of Temik 15G. In 1990 registration of Temik 15G was removed after field tests found residues in potatoes above tolerance levels (U.S. E. P. A., 1990). The American Academy of Pediatrics (1990) reported a child consuming a potato with these levels over time would consume a dose one-tenth of LD<sub>50</sub>, well above toxicity threshold. From 1966 to 1982 there were 165 incidents and several deaths regarding workers exposed to Temik 15G (U.S. E. P. A. 1988). A study involving German greenhouse workers, revealed a decrease in acetylchlolinesterase for up to ten days following exposure (Wagner and Hermes, 1987). In addition to these issues, Temik 15G has been linked to 35% of attempted homicides, 40% of suicides and 10% of accidental poisonings (Ragoucy-Sengler et al., 2000) in the United States population. The product has also been linked to one of the worst chemical disasters since the end of World War II occurring in Bhopal, India (a manufacturing site for methyl isocynate (MIC) used in formulating Temik 15G) in 1984. Sabotage of the plant resulted in a release of a toxic cloud of MIC into the atmosphere causing 5,000 direct deaths and up to 200,000 illnesses including respiratory problems, eye damage, and death to babies in fetal and new born state (Metha, 1990). Because of this string of deaths and

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devastation, EPA began closely scrutinizing product use. From an extensive EPA report (EPA, 2010) showing risks and residue levels of Temik 15G and its metabolites in particular crops, Bayer Crop Science decided to phase out production and distribution of Temik 15G globally by 2018. Temik 15G use in potatoes and citrus was banned in 2012 and all remaining uses end by 2018. In the meantime, new requirements went into effect to change labeling to protect ground water near cotton, soybean and peanut farms (Cone, 2010). Hoewever, due to stipulated EPA requirements, Bayer Crop Science decided to stop production of MIC in 2012 and not later as was first announced (Kirkpatrick et al., 2011). Regardless of safety issues, efficacy of Temik 15G had been on decline. An early study revealed Temik 15G's half life at two months in some fields and eight months in other fields (Jones and Marquardt, 1987). In yet another study, half life was shown to be 408 days (World Health Organization, 1991). However, performance of Temik 15G began to decline in 1998 in Mississippi, Arkansas, Alabama and Louisiana (Lawrence et al., 2004). Lawrence et al. (2004) in a conclusive study across four soil types linked Temik 15G efficacy loss to soil type and degradation by soil microorganisms with complete degradation in 12 days to 43 days depending on soil type. Boozer et al. (2006) further validated this study but extended to include efficacy loss on early season insects. Loss of efficacy was linked to degradation in specific soils due to breakdown by microorganisms not due to acquired resistance by R. reniformis.

## Integrated approaches to growing cotton in *R. reniformis* infested soils

Despite presence of adequate seed treatments, use of multiple management strategies involving crop rotation using grain crops, peanut (*Arachis hypogea*) or reniform resistant soybean (*Glycine max*) cultivars has been encouraged (Windham and Lawrence, 1992; Robinson et al., 1997; 1999; 2001; Koenning et al., 2005). However, rotation only reduces population for one year allowing rapid population increase the following growing season when there is a return to cotton (Davis et al., 2003) still requiring use of a suitable nematicide in future years of *G. hirsutum* production. Other alternatives to nematicides and crop rotation include cover crops and soil amendments and reducing plant stresses that result from compaction and poor drainage (Gaur and Perry, 1991). Another method of addressing suppression of *R. reniformis* is use of host plant resistance since resistance genes have been identified in *G. hirsutum* (Cook et al., 1997 a; Jones et al. 1988; Yik and Birchfield, 1984). Despite gene indentification, incorporation into commercial and elite varieties has proven too difficult. Therefore, integrated approaches have been strongly encouraged to manage nematodes and facilitate yield enhancement. Integrated nematode management programs have many limitations, but have been made necessary with loss of Temik 15G.

# Variety performance and breeding programs involving R. reniformis

There is need for a complete integrated approach that leads to limited reliance on chemicals for *R. reniformis* management. A portion of this integrated approch involves identifying tolerance levels of current cotton germplasms to *R. reniformis*.

Presently, there are no marketed nematode resistant *G. hirsutum* varieties, but a large effort has been directed toward resolving this need. There are varieties that have shown tolerance to nematodes (Usery et al., 2004, and 2005), but most studies only show low to moderate tolerance by currently grown varieties (Starr et al., 2007; Weaver et al., 2007). Gene identification driving nematode resistance in *G. hirsutum* has made positive strides. Davis et al. (2011) reported *M. incognita* resistance is a multi-gene trait difficult

to maintain in breeding programs while Bell and Robinson (2004) reported resistance to R. reniformis requires introgression of genes from Gossypium longicalyx. Robinson and Bell (2006) further reported DNA markers had been identified imparting resistance to M. *incognita* and *R. reniformis*, but, to date, the best industry can hope for is tolerance. The United States Department of Agriculture (USDA) released two cotton varieties (LONREN-1 and LONREN-2) originating from G. longicalyx (a wild Gossypium species from Africa) virtually resistant to R. reniformis (Robinson et al., 2007 a and b; Starr et al., 2007). Percival et al. (1999) and Yik and Birchfield (1984) cited G. longicalyx as having complete resistance, preventing R. reniformis females entering the root from forming their normal kidney shape. This prevents nematode mating, reproduction and egg production reducing subsequent generations. However, G. longicalyx has poor growth habit in spite of being adapted to dry and high saline environments. Bell (1984) reported incompatibility between G. hirsutum (2n=52, similar to other Gossypium sp.) and G. *longicalyx* (2n=26) makes it difficult to successfully cross the species. However, progress has been made relative to this issue (Avila et al., 2005; 2006; 2008; Dighe et al., 2005; Bell and Robinson, 2004; Robinson et al., 2004; Bell et al., 2009; Young et al., 2004; and Robinson et al., (2007), introgressing resistance to R. reniformis into G. *hirsutum* from G. longicalyx.

LONREN cultivars have great susceptibility to root borne fungi and can support only low populations of *R. reniformis* in the greenhouse and field (Bell et al., 2009; Weaver et al., 2011; Weaver et al., 2013) despite having excellent fiber quality. Where *R. reniformis* populations ranged from 10,000 to 50,000 per 100 cm<sup>3</sup> of soil at planting, LONREN lines were intolerant promoting smaller root systems, stunted shoots and reduced yields (Nichols et al., 2010; Sikkens et al., 2011). It has been shown that LONREN lines provide a hypersensitive reaction where root cell tissue damaged upon infection promotes *R. reniformis* death, but negative plant growth still occurs. The negative effect occurs between radical emergence and full seedling growth (Sikkens et al., 2011; Weaver et al., 2013). Schrimsher, et al. (2014) showed nematicides could overcome this brief period of susceptibility.

BARBEN is yet another *Gossypium* species derived from *Gossypium barbadense*. Through years of studies and searching, a number of *G. barbadense* cultivars were discovered to have resistance to *R. reniformis* reducing egg production to as low as 8% and affecting subsequent generations (Yik and Birchfield, 1984; Robinson and Percival, 1997; Robinson et al, 2004). Robinson et al. (2004) found accession GB-713 reduced egg production of *R. reniformis* to as low as 3% and is now being used to introgress resistant genes into *G. hirsutum*. In 2012, USDA, Mississippi State University, and Cotton Incorporated launched BARBEN-713. Sikkens et al. (2012) reported this cultivar supported continuous low levels of *R. reniformis* and yielded comparable to commercially available cotton cultivars indicating a potential for being crossed with high yielding, commercially available germplasms.

Numerous studies have been conducted to evaluate performance of commercial varieties in nematode infested soils. Since 1988, eleven breeding lines tolerant to *M. incognita* have been released (Jones et al, 1988; Cook et al., 1997a; Cook et al., 1997b; Cook and Robinson, 2005). These varieties yield well in *M. incognita* infested fields of their developed production regions. However, according to Koening et al. (2001), these varieties might not be adapted to a wide geographic area. Wheeler, et al. (2014) reported

a positive economic interaction between nematicides plus foliar applications of Vydate C-LV<sup>®</sup> and variety in *M. incognita* populations. Usery et al. (2004; 2005), Legee et al. (2007) and Blessitt et al. (2012) reported several varieties showed tolerance in high R. reniformis infested soils. Usery et al. (2004) reported earlier maturing varieties showed greater tolerance to *R. reniformis* providing higher yields and lower nematode feeding activity in the roots. Further work evaluating commercial variety performance in nematode infested soils was reported by Phipps and Eisenback (2005) and Davis (2005) as it related to *M. incognita* and Sciumbato, et al. (2005) and Blessitt et al. (2012) as it related to R. reniformis. These groups showed no difference in tolerance related to cotton maturity and nematode species. Koenning et al. (2005), however, reported late maturing varieties performed better than early maturing varieties in soils infested with the Columbia lance nematode (Hoploaimus columbus) while Williams et al. (2004) reported similar findings with *M. incognita*. Phipps and Eisenback (2005) further reported net dollar return was significantly greater when using tolerant cotton varieties planted in M. incognita infested fields. They also reported nematicides were still economically beneficial when used with tolerant varieties. Several public sector varieties show promise in highly infested nematode soils (Davis et al., 2010). The only advanced technology (Widestrike/Roundup Flex or BG 2/RF) containing G. hirsutum variety evaluated that has shown nematode tolerance is Phy 367 WRF. Phy 367 WRF showed excellent response in *M. incognita* infested soils despite not being treated with Telone II or Temik 15G (McPherson and Rush, 2011). Today, Fiber Max and Stoneville, subsidiaries of Bayer Crop Science, are discussing potential tolerance to nematodes, especially *R. reniformis*. There is a great need to understand the fruiting mechanisms and performance of the new, high yielding *G. hirsutum* varieties in nematode infested soils. With cost of *G. hirsutum* seed and technology it is important to minimize controllable risks. In addition, new *G. hirsutum* varieties containing advanced technologies have ability to yield approximately 400 lbs. of lint cotton/acre more than the older technology containing varieties due to increased fruit retention (Stewart and Smith, 2007). With yield potentials of 1,400 to 1,600 lbs. lint/acre, all yield hindering events must be minimized. *R. reniformis* tolerant varieties can greatly improve yield and improve NST efficacy.

# Importance of plant mapping monitoring procedures in evaluating cotton development in nematode soils

Understanding *G. hirusutum* growth and development is critical in implementing management strategies for maximizing yields, profits and understanding stress effects. *G. hirusutum* possesses a unique fruiting pattern of simultaneous reproductive and vegetative growth which makes cotton much different in growth pattern for other row crops. This growth pattern makes *G. hirusutum* an ideal plant in which to evaluate and quantify stresses due to nematodes and environment (Jenkins and McCarty, 1995; Kerby et al., 1987; Smith et al., 1996; 1998).

The best method of understanding how a variety fits a system is by understanding its fruiting architecture via plant mapping processes (Jenkins and McCarty, 1995; Kerby et al., 1987; Smith et al., 1996; 1998). Plant maps will determine growth propensity and fruit retention under adverse environmental conditions. In addition, greenhouse evaluations must occur concurrently to establish a tolerant innoculated population (*Pi*) for each variety tested.

Gutherie and Kerby (1993) reported *G. hirusutum* growth maintains a record of its response to environmental conditions and management inputs which can be traced by observing its vegetative structure and fruit distribution. The vegetative and reproductive growth distribution can be quantified by the plant mapping process. Biotic or abiotic stresses can be placed on a developmental time-line by denoting where the symptoms were left on the plant. Early-season conditions are recorded in vegetative growth and square retention levels while mid-season effects are observed in internode lengths and boll retention. Late-season influences impact location of last harvestable boll and degree of secondary growth. The importance of plant mapping has been well documented (Jenkins and McCarty, 1995; McCarty et al., 1994, Albers, 1993; Hake et al., 1990). Inseason plant mapping process has been used extensively to quantify treatment effects in *G. hirusutum*. End-of-season box mapping is an intensive process where every position on the cotton plant is accounted for by boll number, weight by position, contribution by position and cumulation over time relative to yield (Jenkins and McCarty, 1995).

Smith and McCarty (1996) used in-season plant mapping and box mapping to demonstrate effectiveness of Temik 15G applied at-planting and as a side-dress in *G. hirusutum* growing in *R. reniformis* infested soils. From this methodology, they were able to capture treatment fruiting pattern differences, growth differences, maturity and yield differences. Turnage and Smith (1998) further used in-season plant mapping to demonstrate how Temik 15G performed compared to Acephate 15G under heavy thrips pressure across 15 *G. hirusutum* varieties based on fruit retention, height to node ratios, earliness and yield when grown in *R. reniformis* infested soils. The pertinence of early season insect management was also reported using in-season plant maps and box

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mapping techniques (Stewart et al., 2001; Phelps et al., 1996). Using in-season plant mapping and box mapping, Smith and Turnage (1998) further demonstrated how use of Temik 15G benefited an early season cotton variety by reducing thrips damage in R. *reniformis* infested soils. From this data they demonstrated value of maintaining apical dominance and how apical dominance related to yield, earliness, ease of harvest and increased first harvest. Smith et al. (1999) demonstrated efficacy of Bollgard technology across 11 G. hirusutum varieties at 14 locations using in-season plant mapping (prebloom, and 30% open boll). He also used this method to determine timing of defoliation by variety and how harvest of difficult to harvest G. hirsutum varieties could be improved with the use of Finish harvest aid. Presley et al. (1999) used in-season plant mapping to demonstrate fruiting mechanisms in Deltapine Seed Bollgard G. hirsutum varieties compared to experimental varieties in North Delta of Mississippi. In-season plant mapping and box mapping processes were used to quantify fruiting pattern of Roundup Ready varieties treated with labeled and non-labeled applications of Glyphosate prior to the Roundup Ready Flex technology (Monks et al., 2007; Stewart et al., 2005; Pline-Srnic et al., 2004; Viator et al., 2004; File et al., 2000; Jones and Snipes, 1999). Jenkins et al. (1990 a and b; 1990b) and Kerby et al. (1987) using box mapping, reported 66 to 75% of the yield originated from first position fruiting sites on the sympodial (fruiting) branches while 18 to 21% came from second position fruiting sites. Jenkins et al. (1990b) also reported, by use of box mapping across eight G. hirsutum varieties, seed cotton per boll varied among fruiting sites where bolls at the first fruiting position along sympodial branchs were 14% larger than from second positions which was 21% larger than bolls than position three. In this study, Jenkins et al. (1990b) reported boll weights

increased from node 6 to node 12 and declined at upper nodes. Jenkins and McCarty (1995), in the most conclusive box mapping project, reported percentage of mature bolls at harvest began to decline beyond node 15 at fruiting position one and chance of harvesting these positions was reduced at all sites as was fruiting positions  $\geq 2$ . In this study, they further reported higher dollar value bolls were located between nodes 7 and 13 at first position fruiting site in the early maturing variety DES 119. Second positions and >2 positions were lower in value. The later maturing variety, Deltapine 90, showed a higher dollar value between node 8 and 16 at the first position fruiting site. This work indicates importance of maintaining first positions on sympodial branches and that second position fruiting positions can't totally compensate for loss of the first position. Furthermore, the first position is the only fruiting site differentiated in apical meristem (Mauney, 1986). Sadras (1995), in a comprehensive review on G. hirsutum compensation using plant mapping, reported that loss of key fruiting positions could be overcome but depended on plant-water reserves, photosynthesis, changes in plant structure and carbon/nitrogen reserves. In nematode infested soils, this knowledge is pertinent due to root feeding by nematodes and subsequent loss of first position retention sites (Lawrence et al., 1998; 2001; 2002 a). These are very conclusive trials examining fruiting mechanics in nematode infested soils. Davidonis et al. (2004) used plant mapping to evaluate lint quality relative to lint diameter (micronaire) which indicated lower bolls had lower micronaire followed by an increase in the middle fruiting zone followed by a reduction in micronaire at upper nodes. Micronaire is only one quality parameter affectin fiber spinability and marketability.

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There have been several trials showing influence of nematicide treatments on fruiting patterns in conventional and Bollgard/Round-up Ready *G. hirusutum* varieties. Lawrence et al. (1998; 2001; 2002 b) and Lawrence and McLean (2002) showed the influence of nematicide treatments on *G. hirusutum* in *R. reniformis* infested soils via the number of bolls retained per position and lint cotton weights per fruiting position. However, there are few trials (Usery, 2004; 2005) of this nature on new Bollgard II/Roundup Ready Flex (BG2/RF) technology which is expensive, but capable of providing higher fruit retention than observed in the older BG/RR technology and conventional varieties (Stewart and Smith, 2007). This increased boll retention and maintenance to harvest mandates adequate root development to enhance complete nutrient and water uptake to maximize yield.

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#### CHAPTER III

# EFFECTS OF SEED TREATMENT NEMATICIDES WITH AND WITHOUT FOLIAR APPLICATIONS OF VYDATE C-LV<sup>®</sup> ON THE GROWTH AND DEVELOPMENT OF *GOSSYPIUM HIRSUTUM* GROWING IN *ROTYLENCHULUS RENIFORMIS* INFESTED SOILS

#### Abstract

Reniform nematode (*Rotylenchulus reniformis* Linford and Oliveira) currently infests 36% of the Mississippi cotton acreage and causes economic losses of \$130 million annually. With Temik 15G being removed from the market and high expense of soil fumigants, there is a need to develop an integrated nematode management program centering around Nematicide Seed Treatment (NST) with and without foliar applications of Vydate C-LV<sup>®</sup>. In greenhouse studies, all NSTs showed greater root and shoot weights compared to the untreated control (UTC). Aeris<sup>®</sup> + Votivo<sup>®</sup> produced greater root and shoot weights in inoculated populations (*Pi*) up to 5,000 reniform nematodes/500 cc of soil. Relative to root and shoot growth, Aeris<sup>®</sup> treated plants began having less growth at 2,500 reniform nematodes/500 cc. Temik 15G increased shoot weights until *Pi* of 7,500 reniform nematodes/500 cc of soil, but root weights of Temik 15G treated plants at all nematode levels were not better than the UTC at 0 and 2,500 reniform nematodes/500cc of soil respectively indicating root growth restriction from Temik 15G not observed in NSTs. In-season plant mapping indicated Node of First

Fruiting Branch (NFFB) was reduced with all nematicide treatments. Plant height and height to node ratios (HNR) were increased by the addition of Vvdate C-LV<sup>®</sup> treatments above the NSTs alone, as shown by accumulated internode measurements, while all nematicide treatments improved growth over the UTC. During square growth period, no retention differences occurred at fruiting position one but Vydate C-LV® treatments provided higher retention at positions greater than (>) two. Final fruit evaluation indicated no difference in retention at position one, but Vydate C-LV<sup>®</sup> treatments did increase retention at position two. From a plant zone perspective, little difference in zone one (Nodes 5-9) and zone two (Nodes 10-14) existed during square. However, in-bloom retention began to improve in all nematicide treatments treated with Vydate C-LV® across all zones for position one while at position two the Vydate C-LV<sup>®</sup> treatment increased retention at zones one. At position two and three (N 15-19) during open boll growth phase at fruiting position one and two. All NSTs were improved using Vydate C-LV<sup>®</sup> equaling that of Temik 15G alone, but Temik 15 G still provided greater boll retention in *R. reniformis* infested soils. This was further observed in yields where all treatments increased yield over the UTC and Vydate C-LV<sup>®</sup> treatments increased yields above the NST alone treatments.

Key study nematicides: Nematicides used in the seed treatment study included the following; \*Temik 15 G (Aldicarb): 2-methyl-2-(methylthio)-propionaldehyde 0methylcarbanoyloxime); \*Vydate C-LV<sup>®</sup> (Oxamyl): methyl N'N'-dimethyl-N-[(methyl carbamoyl) oxy]-1-thiooxamimidate; \*Aeris<sup>®</sup> (Thiodicarb): dimethyl N,N'-[thiobis[(methylimino) carbonyloxy]bis [ethanimidothioate]. Votivo<sup>®</sup> (*Bacillus firmus*).

#### Introduction

The reniform nematode (*R. reniformis* Linford and Oliveira), a plant parasitic nematode, has become the most damaging G. hirsutum pathogen. R. reniformis, first described in 1931 (Linford and Oliveira, 1940), is a tropical and subtropical pest present throughout the United States G. hirsutum producing region (Heald and Robinson, 1990; Kinloch and Sprenkel, 1994; Starr, 1998; Koenning et al., 1999). Since 1960, R. *reniformis* has shown an adaptive capability to survive colder environments allowing colonization of much of the eastern half of the G. hirsutum belt (Heald and Robinson, 1990) and as far north as Lubbock, Texas and the Missouri bootheel (Heald and Thames, 1982; Wrather et al., 1992). Today, R. reniformis has been identified and associated with a 7% annual G. hirsutum yield loss totaling nearly \$126 million in Mississippi, Alabama, Tennessee, Texas, Missouri, Florida, North Carolina, Louisiana, South Carolina, Arkansas and Georgia (Blasingame and Patel, 2011; Koenning et al., 1999). In Mississippi, an annual yield loss of 235,398, 252,023 and 56,378 bales occurred in 2004, 2005 and 2011, respectively (Blasingame, 2004; 2005; Blasingame and Patel, 2011). By 2002, more than 32% of the G. hirsutum acres in Mississippi were infested with R. reniformis causing a 5.5% yield reduction (Lawrence and McLean, 2002). Gazaway and McLean (2003) reported R. reniformis infested more than 36% of Alabama G. hirsutum acreage and was increasing.

Since 2004, the cotton industry began moving away from the granular, at-planting treatment with Temik 15G for nematode management. Prior to this time, and for more than 40 years, Temik 15G was the main-stay for nematode management in the cotton industry. However, in 2012, Bayer Crop Science made the decision to cease production

of this product, and Nematicide Seed Treatments (NSTs) have replaced Temik 15G in the industry. Padgett et al (2004) reported some NST treatments were as effective as Temik 15G applied at its nematicide rate of 0.75 lbs ai/Ac and reduced galling over the untreated control (UTC), but did not improve maturity or yield indicating lack of longevity compared to Temik 15G. Kirkpatrick and Monfort (2004) reported NSTs did not differ from Temik 15G 14 to 35 days after planting (DAP). In addition, they reported NSTs applied at 100 g ai/kg of seed was similar to Temik 15G applied at 0.75 lbs ai/Ac. Monfort et al. (2004) reported Meloidogyne incognita (root knot nematode) numbers and gall numbers were also reduced using NSTs similar to Temik 15G. A major concern of using NST was lack of the early season insect control obtained with Temik 15G. Brown et al. (2008) reported tobacco thrips (Frankliniella fusca) damage reduced early cotton root growth and yield in *R. reniformis* infested soils. However, the group did not evaluate the loss of cotton plant maturity as a result of the combined effects from the nematodes and thrips. This research lead the industry to combine seed treatments containing insecticidal modes of action with those of nematicidal modes of action or mandating an over-top application for insect management to maintain normal cotton growth.

Aeris<sup>®</sup> Seed Applied System (imidachloprid + thiodicarb at 0.825 mg ai/kg of seed) is listed for suppression of *M. incognita* and *R. reniformis* as well as *F. fusca*, soybean thrips (*Sericothrips variabilis*), onion thrips (*Thrips tabaci*) and cotton aphids (*Aphis gossypii*). There is an additional option of using TRILEX<sup>®</sup> (seed treatment fungicide) for control of *Rhizoctonia*, *Pythium*, *Thielaviopsis* and *Fusarium* added in this mixture. Graham et al. (2007) reported AERIS<sup>®</sup> Seed Applied System increased plant

height, leaf area, white-bloom count and yield when compared to Avicta<sup>™</sup> which was similar to findings by Kemerait et al. (2006; 2007; 2008). Kemerait et al. (2007; 2008) reported Temik 15G provided better yields and return on investment when compared to NSTs in nematode infested soils.

Extension of *R. reniformis* management beyond 35 days after planting has been made possible with foliar applied Vydate C-LV<sup>®</sup> (Lawrence and McLean, 2000; 2002; 2003). Vydate C-LV<sup>®</sup> with nematicide/insecticide properties remains a viable tool in managing cotton nematodes because of the ease of foliar application and phloem transmission to the root system (Hsu and Kleier, 1996). This tool becomes crucial since *R. reniformis* obtains maximum population densities at a time cotton is in its peak reproductive phase (Lawrence and McLean, 1995 a and b; 1996 a and b). Parasitism by *R. reniformis* results in reduced cotton yields and requires additional treatments beyond the control obtained by the NSTs.

The objectives of this study were to determine if NSTs provide adequate *R*. *reniformis* suppression to maintain fruiting architecture in cotton varieties compared with Temik 15G; to determine if foliar applications of Vydate C-LV<sup>®</sup> enhances cotton fruit retention where NSTs are used in *R. reniformis* soils and to determine effect of the NST on cotton plant growth and maturity. Further exploration via additional greenhouse studies were used to determine how *R. reniformins* population affects growth of cotton treated with the NSTcompared to Temik 15G.

#### **Materials and Methods**

#### In-field nematicide study

Studies were conducted at two locations, Tennessee Valley Research and Extension Center (TVREC) of Auburn University (AU) in Belle Mina, Ala. and R. R. Foil Plant Science Research Center of Mississippi State University (MSU) in Starkville, Miss.

Treatments consisted of two NSTs (Aeris<sup>®</sup> at 0.075 mg ai/seed rate and Aeris<sup>®</sup> + Votivo<sup>®</sup> at 0.424 mg ai/seed rate) (Bayer Crop Science-Raleigh, NC) and one in-furrow, at-planting treatment (Temik 15G at 0.75 lbs ai/ac) to evaluate effect of at-planting applications without post-planting application of Vydate C-LV<sup>®</sup> (Dupont USA-Wilmington, DE) (Table 3.1). Additional treatments included previous treatments with a post-plant foliar application of Vydate C-LV<sup>®</sup> at 0.24 lbs ai/ac at sixth true leaf growth stage. A second application of Vydate C-LV<sup>®</sup> was applied ten days later. NSTs without Vydate C-LV<sup>®</sup> received insecticide Orthene (acephate) 90S<sup>®</sup> at 0.75 lb ai/Ac (Table 3.1). Continued insect management was conducted similarly on an as needed basis applied with a pre-calibrated ground driven sprayer. Vydate C-LV<sup>®</sup> and Orthene 90S<sup>®</sup> treatments were applied with a CO<sub>2</sub> back-pack sprayer calibrated to deliver 15 gallons water per acre. Phy 375 WRF (Dow AgroScience-Zionsville Road Indianapolis, IN) was the Gossypium hirsutum variety used. Planting was conducted on May 1, 2012 and May 15, 2012 at TVREC and MSU, respectively, using a four-row Almaco cone planter (Allan Manufacturuing Company, Nevada, IA). Weed control consisted of applications of Power Max<sup>®</sup> (glyphosate) (Monsato-St. Louis, MO) over-the-top of cotton at 1.0 lb ai/Ac followed by a lay-by application of Karmex DF <sup>™</sup> (diuron) (DuPont USA-Wilmington,

DE) at 1.0 lb ai/Ac. Soil tests were conducted prior to planting at both locations and analysis processed at Mississippi State University Extension Soil Testing Lab (Mississippi State, Miss.). Soil type at TVREC was a Decatur silty, clay loam while the MSU location was a Marrietta fine, sandy loam. Both locations had irrigation with MSU location having furrow irrigation and TVREC having center pivot irrigation. Due to dry weather, TVREC was the only location irrigated.

#### Experimental design and trial establishment

Trial design used at both locations was a randomized complete block (RCB) design consisting of five replications at MSU and four replications at TVREC. This statistical method was selected to address the spatial distribution of nematodes across the field thereby reducing variability of nematode populations existing between plots. Data were analyzed with Analysis of Variance (ANOVA) for a RCB (ARM 8 statistical software-Gylling Data Management, Brookings, SD) where block and treatment effects were evaluated to minimize dgree of error and improve confidence intervals among experimental units. Means were separated using Least Significant Difference (LSD) at 0.05 probability level. Plots consisted of two-rows 50.0 feet at MSU and 25 feet at TVREC long with 10.0 foot alleys. Row spacing consisted of solid planting patterns of 40.0 inch at TVREC and 38.0 at MSU with a seeding rate of 4.0 seed per foot of row. Seed was pre-counted before planting using a Model U Seed Counter (International Marketing and Design Corporation, San Antonio, TX). Border effects were reduced by planting sides with additional cotton and using a solid planting pattern. Lack of bordering in front and back of trial area was compensated for by acquiring samples from within plots to avoid edge effects.

#### In-season evaluation prior to fruiting

#### Evaluation of vigor, plant population and hypocotyl lengths (")

Visual plant vigor and plant populations were evaluated at 14 days following emergence. Vigor was determined using two processes: 1. Visual assessment on a scale of one to five where one had greatest plant growth and five the worst and, 2. hypocotyl measurement. Hypocotyl measurement involved measurement of length from seed embryo axis to cotyledonary node. Hypocotyl measurements, as opposed to visual evaluations, provide a quantifiable and accurate method to analyze vigor (Legee and Smith, 2002). Plant population was determined by counting every plant in all plots to determine plants per acre.

### In-season evaluation during fruiting

#### **Evaluation during mid-square growth stage**

An extensive plant mapping program during reproduction was conducted where six consecutive plants having a normal terminal were destructively sampled and measured via plant mapping processes for boll retention and growth (Gutherie and Kerby, 1993). This process included three evaluation timings; mid-square, bloom and the open boll growth stages.

At mid-square, evaluation criteria included: plant height (PH) (from the cotyledonary node to the terminal), node of first fruiting branch (NFFB), total nodes (TN) (cotyledonary node was treated as zero), height to node ratio (HNR) (determined by dividing total plant height by total nodes), retention by fruiting position along sympodial (fruiting branch) and by zone and average plant height by node measurements. Fruiting zones were established up the cotton plant main axis based on node numbers where zone

1 represented nodes 5-9, zone 2 represented nodes 10-14, zone 3 represented nodes 15-19 and zone 4 represented nodes greater than 20. Average plant height by node measurements was conducted by measuring each internode length separately from cotyledons to 0.5 inch wide terminal leaf in a manner where overall length culminated in final height (Kerby, et al., 2003). This method facilitated measuring individual internode growth across time as affected by nematicide treatment and *R. reniformis*. Six consecutive plants possessing a normal terminal were sampled destructively per plot providing a total of 30 plants sampled at MSU and 24 plants at TVREC.

#### **Evaluation during bloom growth phase**

Evaluation criteria on six consecutive plants per plot during late bloom included: PH in inches, TN, HNR, nodes above white flower (NAWF), node of white flower (NOWF), retention by position and by zone and average plant height by node measurements conducted at TVREC (data was lost from MSU location). In addition, caliper (General Ultra Tech, Port Washington, NY) readings were taken at the cotyledonary node to obtain basal stalk diameter and from unopened first position bolls at node 9 and 12 from the terminal to determine treatment effects on boll growth.

# Evaluation during open boll growth phase

Evaluation during open boll plant growth phase on six plants destructively sampled at about 30% open boll within the earliest treatment included: PH, TN, cumulative plant height, node above cracked boll (NACB), fruit retention by position and by zone and percent open boll.

## Machine harvest

Defoliation was conducted based on visual assessments of 60% open boll with harvest aids applied using high clearance ground equipment. Harvest was conducted using a John Deere 9965 (Moline, IL) small plot machine harvester equipped with a Rice Lake 9201i weighing system (Rice Lake Weighing Systems-Rice Lake, WI) to measure seed cotton of individual plots on-the-go. Seed cotton weights were converted to lint pounds per acre using lint percentages established via University Official Variety Trials at Mississippi State University (Mississippi State, MS).

# R. reniformis sampling and processing

*Rotylenchulus reniformis* collection included nematode soil samples collected prior to planting from each plot to establish an initial population density. Nematode populations were monitored at-planting, square, bloom and open boll. Core soil samples were acquired using a fluted probe designed to collect multiple samples per plot. Probe dimensions were 3.44 inches at the top and tapering to 0.75 inches at the bottom facilitating multiple samples without soil loss. Length of sample device was 11.0 inches to guarantee acquisition of 500 cc of soil. Samples were acquired from the side of the emerged row at a distance of about six inches in zig-zag pattern allowing six samples to be acquired at three samples per row. Sampling was conducted at an approximate depth of four inches. The sampling process was always conducted when the soil possessed adequate moisture levels, preferably at field capacity since *R. reniformis* move deeper into the soil profile as soil dries. Samples were bagged in plastic bags and kept in cold storage until extraction using a semi-automatic elutriator (W.S. Tyler Co, Mentor, OH) and centrifugal (1 EC Model K Centrifuge, Needham Hts, MA) flotation (Byrd et al., 1976). *R. reniformis* extraction process was as follows: Collected soil from individual plots was placed into a 450 ml beaker and processed through a 60 mesh screen followed by a 400 mesh screen using an aqueous extraction process of the elutriator. Soil was removed and placed into a 250 ml beaker, water drained and sample poured into centrifuge tubes where it contained 10 to 15 grams of 1.0 inch of soil and spun for six minutes at 2,500 RPM. Excess water was removed and mixed with a sucrose mixture (454 g sucrose per 1,000 ml of water) to the top of the centrifuge tube and followed by a one minute process in the centrifuge. The liquid was poured through a 500 mesh screen and sample refrigerated until counted. The resulting nematodes were enumerated using a stero-microscope (Nikon AFX-11A, Minato-ku, Tokyo).

# Effects of *R. reniformis* nematodes under greenhouse environments on *G. hirsutum* development treated with nematicides

# Trial establishment and experimental design

Two seperate greenhouse studies were established using the cotton variety Phy 375 WRF planted at two seeds per 3.0 inch clay pot into an autoclave, fine sandy loam. All pots were brought to the same level to ensure 500 cc. Planting depth for all seed was 0.5 inch. Upon emergence, one plant was removed to leave one plant per container. Treatments included Temik 15G at an equivalent rate of 0.75 lbs ai/Ac, Aeris<sup>®</sup> at 0.075 ai/seed rate and Aeris<sup>®</sup> + Votivo<sup>®</sup> at 0.424 mg ai/seed rate (Table 3.2). Nematode populations were applied in a liquid solution using a graduated pipette and included 0, 2,500, 5,000, or 10,000 *R. reniformis* per 500 cc of soil. Each study was conducted for 90 days. Experimental design was a RCB design using four replications. Data were analyzed via the ANOVA for a RCB (ARM 8 statistical software-Gylling Data

Management, Brookings, SD) where block and treatment effects were evaluated to minimize dgree of error and improve confidence intervals among experimental units. Means were separated using Least Significant Difference (LSD) at 0.05 probability level.

#### **Evaluation criteria**

Before harvest evaluations included TN, PH, NFFB, HNR and basal stalk diameter. At harvest evaluations included root and shoot biomass and nematode extraction (eggs and juveniles). At harvest, shoot biomass was separated from the root biomass using hand pruners. The shoot was then weighed and recorded. Roots were extracted from soil in a bucket. Soil-free roots were soaked in a 10% bleach solution and stirred in solution for three minutes and roots weighed. The remaining solution was poured through 250 over 500 mesh screen to obtain eggs. Remaining soil was mixed with 1,000 ml of water and processed through a 60 over 325 mesh screen to obtain juvenile numbers and centrifuged for six minutes at 2,500 rpm. Excess water was removed and mixed with sucrose mixture (454 g sucrose per 1,000 ml of water) followed by a one minute centrifuge process at 2,500 rpm. The liquid was poured through a 500 mesh screen and sample refrigerated in a 250 ml beaker until counted. Nematode numbers were surveyed via stereo microscope for *R. reniformis* juveniles and eggs by pipetting 20 mls of liquid into a quadrated petri dish.

#### **Results and Discussion**

#### In-the-field evaluation of *R. reniformis* populations across time

In field evaluation of cotton using plant mapping processes is an in-depth process that generates accurate growth and development data relative to the effects of a treatment under specific stresses like *R. reniformis* (Gutherie and Kerby, 1987; Jenkins et al., 1995; 1990 a; 1990b; Smith et al., 2003; 1999; 1998; 1996). This coupled with detrimental effects from *R. reniformis* and its seasonal population progression, allows for accurate monitoring of growth.

*R. renifor*mis population progression across time becomes important in determining impact on growth and development of G. hirsutum at each growth stage. Further relating nematode numbers to root development has established effective treatment against R. reniformis nematode populations resulting in greater root development at season end (Lawrence and McLean, 1995 a and b; 1996 a and b). Rotylenchulus reniformis populations in 500 cc of soil at MSU location showed low populations at planting that continued into square period (Table 3.3 and Fig. 3.1). These numbers tended to increase across all treatments during bloom evaluation. This trend was similar to Lee et al. (2015) where *R. reniformis* populations in fields of continuous cotton had the lowest population during the spring but increased steadily during the season. Rotylenchulus renoformis numbers during open boll growth stage where all nematicide treatments and nematicides followed by applications of Vydate C-LV<sup>®</sup> were higher than untreated. *Rotylenchulus reniformis* populations at TVREC were higher at planting than at the MSU location, and this trend continued throughout the growing season. During square (40 DAE), nematode numbers were higher in all nematicide treatments compared to untreated while all nematicide treatments followed by Vydate C-LV<sup>®</sup> became greater in population than NSTs without Vydate C-LV<sup>®</sup>. Temik 15G was not different from NSTs with or without Vydate C-LV<sup>®</sup>. During bloom (70 DAE) and open boll growth (100 DAE) stages at TVREC (Table 3.4 and Fig. 3.1), nematicide

treatments containing Vydate C-LV<sup>®</sup> applications, with exception of Temik 15G and Vydate C-LV<sup>®</sup>, were higher in *R. reniformis* numbers than NST with no Vydate C-LV<sup>®</sup>. *R. reniformis* numbers trended lower between square and open boll.

### In-the field evaluation prior to fruiting of cotton in *R. reniformis* infested soils Effect of nematicides on vigor, plant population and hypocotyl length

Higher plant population occurred in the untreated compared to the nematicide treatments at MSU with having Aeris<sup>®</sup> the highest plant population of nematicide treatments (Table 3.5). No differences in plant population occurred at TVREC (Table 3.5) location.

Vigor at both locations increased with all nematicide treatments compared to untreated. Temik 15G yielded greatest vigor level at MSU location while Temik 15G and Aeris<sup>®</sup> + Votivo<sup>®</sup> yielded highest vigor at TVREC. This was further manifested in hypocotyl length where all nematicide treatments possessed greater hypocotyl lengths than the untreated at both locations (Table 3.5).

# In-the-field evaluation of cotton development across time in *R. reniformis* infested soils

#### **Effect of nematicides on NFFB**

Node of first fruiting branch at both locations was reduced 9-18% for nematicide treatments at MSU location while Temik 15G or Aeris<sup>®</sup> and Votivo<sup>®</sup> reduced NFFB by 8-13%. Of the nematicide treatments Aeris<sup>®</sup> + Votivo<sup>®</sup> and Aeris<sup>®</sup> were greater in NFFB when compared to Temik 15G (Table 3.6) at the MSU location while all nematicides were similar at TVREC (Table 3.8).

#### Effects of nematicides on plant height

Plant height during square was visually observed to trend lower at TVREC compared to MSU due to heavy thrips pressure and cold temperatures after emergence (Table 3.8). After one application with Vydate C-LV<sup>®</sup>, plant height was greater at both locations when compared to untreated plants. Plants with Vydate C-LV<sup>®</sup> applications were similar in height when compared to plants treated with Temik 15G, regardless of location. These results differ from Lawrence and McLean (2000; 2002; 2003). For nematicide treatments without Vydate C-LV<sup>®</sup>, plant heights were similar for the MSU location during square. However, plants treated with Temik 15G were taller compared to plants treated with Aeris<sup>®</sup> or Aeris<sup>®</sup> + Votivo<sup>®</sup> at TVREC (Tables 3.7 and 3.8).

During open boll growth stage, plants treated with Vydate C-LV<sup>®</sup> continued to be shorter when compared to untreated plants, regardless of location (Tables 3.7 and 3.8) At the MSU location, plants treated with Temik 15G had greater height compared to plants treated with Aeris<sup>®</sup>, but not when compared to plants treated with Aeris<sup>®</sup> + Votivo<sup>®</sup> (Table 3.7). At TVREC, there were no height differences between plants treated with Temik 15G, Aeris<sup>®</sup> or Aeris<sup>®</sup> + Votivo<sup>®</sup> (Table 3.8).

Plant height was greater at both locations for all nematicide treatments compared to untreated (Tables 3.7 and 3.8). In addition, at the MSU location, Vydate C-LV<sup>®</sup> applications increased plant height compared to NST alone which was in agrrement with previous literature (Lawrence and McLean, 2000; 2002; 2003). Height of NST plants measured at open boll at the MSU location benefited from two applications of Vydate C-LV<sup>®</sup>.

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#### Effects of nematicides on total node number

Node number at square increased across all nematicide treatments by addition of Vydate C-LV<sup>®</sup> with exception of Aeris<sup>®</sup> + Vydate C-LV<sup>®</sup> at TVREC (Table 3.8). This further enhances the value of Vydate C-LV<sup>®</sup> in improving *R. reniformis* management as previously observed in findings by Lawrence and McLean (1995 a and b; 1996 a and b). Nematicide treatments at both locations without Vydate C-LV<sup>®</sup> increased total node number except for Aeris<sup>®</sup> at MSU (Table 3.7) but Aeris<sup>®</sup> and Aeris<sup>®</sup> + Votivo<sup>®</sup> had similar numbers when compared to untreated plants (Table 3.8). In general, NST treated plants with addition of Vydate C-LV<sup>®</sup> had a node number greater than plants treated with Temik 15G at the MSU location.

During bloom, all nematicide treatments resulted in increased total number of nodes compared to untreated, regardless of location. Vydate C-LV<sup>®</sup> at the MSU (Table 3.7 and 3.8) location increased or had similar node numbers compared to Temik 15G. Aeris<sup>®</sup> + Votivo<sup>®</sup> + Vydate C-LV<sup>®</sup> did not differ from Temik 15G or Aeris<sup>®</sup> + Votivo<sup>®</sup> alone. Total node number was similar or better than Temik 15G for all treatments, regardless of location.

At open boll (final evaluation), untreated plants at TVREC had greater node number compared to NSTs resulting from delayed fruit initiation (Mauney, 1986). At MSU, only plants treated with Aeris<sup>®</sup> had more total nodes than the untreated.

#### Effects of nematicides on Height to Node Ratios

Height to node ratios (HNR) of all nematicides were improved by final evaluation compared to untreated regardless of location (Tables 3.7 and 3.8). The addition of Vydate C-LV<sup>®</sup> made NSTs similar or better when compared to Temik 15G. During bloom no differences occurred at MSU location (Table 3.7) but all nematicides had greater HNR compared to untreated (Table 3.8). Addition of Vydate C-LV<sup>®</sup> resulted in comparable HNR to Temik 15G, similar to Lawrence and McLean (2000; 2002; 2003).

#### Effects of nematicides on average plant height by node

Evaluation of plant height by node is a method by which stress effect can be quantified and identified via internode elongation (Kerby et al., 2003). Average plant height by node at MSU (Table 3.9) location indicates no differences among nematicides until node 13 during square evaluation phase. Temik 15G with foliar applications of Vydate C-LV<sup>®</sup> at node 13 and 15 had greater internode elongation than untreated, but did not differ from Temik 15G treatment or NSTs. Plants treated with Temik 15G + Vydate C-LV<sup>®</sup> had internodes that were about two inches longer than untreated plants at nodes 13 and 15. Internode growth trends at TVREC (Table 3.10) under higher *R. reniformis* populations and early season stress is different from MSU location during bloom (Table 3.9). Differences in internode elongation began at node 1, where plants treated with Vydate C-LV<sup>®</sup> had greater internode elongation compared to untreated. Application of Vydate C-LV<sup>®</sup> also improved internode elongation in plants treated with Temik 15G and Aeris<sup>®</sup> or Aeris<sup>®</sup> + Votivo<sup>®</sup>. Addition of Vydate C-LV<sup>®</sup> increased internode length when compared to Temik 15G indicating NSTs were more beneficial than Temik 15G (Tables 3.9 and 3.10). Addition of Vydate C-LV<sup>®</sup> resulted in elongation at all other nodes when compared to plants not treated with Vydate C-LV<sup>®</sup> or the untreated.

During bloom at TVREC (Table 3.11), all nematicide treatments had greater internode length than untreated plants at node 1, and by node 3, nematicide treatments began to differ in effect. Plant height at node 3 was similar for all treatments except Aeris<sup>®</sup> and untreated plants. At node 5, treatments including Vydate C-LV<sup>®</sup> had greater height than Aeris<sup>®</sup> or untreated. At node 15, plants treated with Vydate C-LV<sup>®</sup> had greater elongation than plants not treated with Vydate C-LV<sup>®</sup> and all were greater than untreated. Nematicides continued to increase elongation for nodes 17-21 compared to untreated.

#### Percent retention across sympodial fruiting positions as affected by nematicides

Percent (%) square retention by position 40 DAE revealed no differences regardless of location, compared to untreated at fruiting position one indicating R. reniformis did not induce fruit loss during square at position one (Schubert et al, 1986). A high degree of retention at this fruiting site is vital in maintaining high yields and quality since it is initiated in the terminal and receives photosynthates from the main axis and subtending leaf as opposed to the fruiting sites farther out the sympodial (fruiting) branch which are nourished primarily by their subtending leaf (Jenkins et al., 1995; Sadras, 1995). At fruiting position 2, all treatments had improved retention compared to untreated plants, but only Temik 15G + Vydate C-LV<sup>®</sup> demonstrated improvement compared to Temik 15G or Aeris<sup>®</sup> at MSU (Table 3.12). Under higher *R. reniformis* population at TVREC (Table 3.13), Vydate C-LV<sup>®</sup> improved retention compared to all treatments except Temik 15G. Fruiting position >2 did not exist at TVREC due to high levels of *R. reniformis*, thrips and cold weather, but the later planted MSU location did have retention at position >2. At MSU location, all treatments had fruit retention greater than untreated and Vydate C-LV<sup>®</sup> improved retention in NSTs and Temik 15G although Temik 15G was similar to Aeris<sup>®</sup> + Vydate C-LV<sup>®</sup> + Votivo<sup>®</sup> (Table 3.12).

During bloom carbohydrate partitioning becomes important as boll development occurs and is partly related to healthy root development prior to bloom (Schubert et al., 1986; McMichael, 1986). At MSU, fruit retention at position one during bloom was greater for all nematicides compared to untreated (Table 3.12) which becomes important since position one produces high quality bolls (Jenkins et al., 1995; Sadras, 1995). Addition of Vydate C-LV<sup>®</sup> only improved retention for the Temik 15G treatment. Retention levels for Aeris<sup>®</sup> and Aeris<sup>®</sup> + Votivo<sup>®</sup> were not impacted by addition of Vvdate C-LV<sup>®</sup>. Nematicides also improved retention at fruiting position one compared to untreated plants at TVREC (Table 3.13) location. Vydate C-LV® treated plants at MSU had higher % retention at fruiting position two compared to all other treatments except Temik 15G, which was similar. Percent retention at fruiting position two was greater than untreated plants at TVREC. Nematicide seed treatments alone had lower fruit retention than Temik 15G at either location for this fruiting position which indicates addition of Vydate C-LV<sup>®</sup> had a greater effect in the plant. Only Aeris<sup>®</sup> and Aeris<sup>®</sup> + Votivo<sup>®</sup> had higher fruit retention than untreated at MSU. For the TVREC location at position >2, only Temik 15G with or without Vydate C-LV<sup>®</sup> retained more fruit than the untreated.

Fruit retention during open boll growth stage at MSU location (Table 3.12) indicates nematicides improved retention at fruiting position one compared to untreated plants facilitating yield improvement (Jenkins and McCarty, 1995; Sadras, 1995). However, under higher *R. reniformis* populations of TVREC (Table 3.13), position one fruit retention with NSTs without Vydate C-LV<sup>®</sup> did not differ from untreated and had lower fruit retention compared to Temik 15G. This could be due in part to a higher *R*.

*reniformis* population and/or early season stress at TVREC. At both locations, addition of Vydate C-LV<sup>®</sup> maintained or improved retention for all treatments. At the MSU location, Aeris<sup>®</sup> + Vydate C-LV<sup>®</sup> was the only Vydate C-LV<sup>®</sup> treatment to exhibit reduced retention compared to Temik 15G. However, addition of Vydate C-LV<sup>®</sup> resulted in greater fruit retention compared to Temik 15G at TVREC.

Percent retention at position two was greater across all nematicide treatments compared to untreated plants at MSU location. At TVREC, fruit retention was improved in all treatments except Aeris<sup>®</sup> + Votivo<sup>®</sup>. At both locations, adding Vydate C-LV<sup>®</sup> in management of *R. reniformis* improved retention of position two fruiting sites across all nematicides.

During open boll, fruiting position >2 retention at MSU only Aeris<sup>®</sup> + Votivo<sup>®</sup> and Aeris<sup>®</sup> + Vydate C-LV<sup>®</sup> had higher retention compared to untreated indicating normal termination on-going within the plant (Jenkins and McCarty, 1995). Aeris<sup>®</sup> + Votivo<sup>®</sup> and Aeris<sup>®</sup> + Vydate C-LV<sup>®</sup> treatments had greater retention compared to the control at this fruiting position indicating delayed maturity resulting from a different architecture. Foliar applied Vydate C-LV<sup>®</sup> treatments with Temik 15G and Aeris<sup>®</sup> + Votivo<sup>®</sup> had similar fruit retention compared to untreated at position >2. Temik 15G + Vydate C-LV<sup>®</sup> had lowest retention at this position. Temik 15G or Aeris<sup>®</sup> + Vydate C-LV<sup>®</sup> resulted in an increase in retention at this fruiting position compared to all other nematicide treatments or untreated. All nematicide treatments had greater retention than untreated plants. Use of Vydate C-LV<sup>®</sup> enhanced performance of NSTs, especially under high *R. reniformis* populations.

## Percent retention of sympodial positions within fruiting zones as affected by nematicides

Percent retention during square (Tables 3.14 and 3.15) for fruiting position one in Zone one (Nodes 5-9) showed no differences for either location. In Zone one at MSU, Vydate-CLV<sup>®</sup> treatment improved fruit retention of Temik 15G and Aeris<sup>®</sup> + Vydate C-LV<sup>®</sup> compared to the untreated at fruiting position two making retention comparable to Temik 15 G. For TVREC addition of Vydate C-LV<sup>®</sup> improved retention compared to untreated. Impacts from Vydate C-LV<sup>®</sup> treatment were evident at TVREC (Table 3.15) with improved fruit retention at position two compared to the control. These impacts were also observed at position > 2 at MSU when Vydate C-LV<sup>®</sup> was combined with Aeris<sup>®</sup> or Aeris<sup>®</sup> + Votivo<sup>®</sup> (Table 3.14), but there was no position > 2 at TVREC. Fruit retention at Zone two (Nodes 10-14) had no differences between nematicide treatments at fruiting position one at MSU, but at TVREC, nematicide treatments had greater fruit retention than untreated plants. In Zone two at fruiting position two, treatment with Vydate C-LV<sup>®</sup> resulted in greater fruit retention compared to untreated, regardless of location. At fruiting position >2 in Zone two,  $Aeris^{\text{(B)}} + Vydate C-LV^{\text{(B)}}$  had greater fruit retention compared to the untreated or any other treatment at MSU, but all treatments of TVREC had greater fruit retention compared to the untreated. In Zone two at fruiting position two at MSU, the only treatment different from untreated plants was Temik 15G + Vydate C-LV<sup>®</sup>. At TVREC, Temik 15G + Vydate C-LV<sup>®</sup> and Aeris<sup>®</sup> + Votivo<sup>®</sup> + Vydate C-LV<sup>®</sup> had greater fruit retention compared to Temik 15G or untreated.

Vydate C-LV<sup>®</sup> treatments, during bloom (Table 3.16 and 3.17) at Zone one, position one, did not improve fruit retention of NSTs compared to their non Vydate C-LV<sup>®</sup> equivalent at MSU (Table 3.16). Temik + Vydate C-LV<sup>®</sup> and Aeris<sup>®</sup> + Votivo<sup>®</sup> +

Vydate C-LV<sup>®</sup> had greater fruit retention at position one in Zone one compared to untreated. In this zone and position at TVREC (Table 3.17), all treatments had greater fruit retention than untreated plants. Vydate C-LV<sup>®</sup> applications at MSU location, enhanced fruit retention at position two across all NSTs making them similar to Temik 15G. Aeris<sup>®</sup> + Votivo<sup>®</sup> and Aeris<sup>®</sup> did not differ from untreated plants in fruit retention in Zone one, position two (Table 3.16). TVREC location followed a similar pattern for position two fruit retention, but Vydate C-LV<sup>®</sup> did not improve Aeris<sup>®</sup> retention at this zone and position. At fruiting position > 2 in Zone one there were few differences compared to untreated plants. At TVREC, all treatments improved fruit retention compared to untreated, but addition of Vydate C-LV<sup>®</sup> to NST's did not improve their performance compared to Temik 15G (Table 3.17). In zone two at position one, all treatments had greater fruit retention than untreated plants at MSU, but Aeris<sup>®</sup> was similar to untreated at TVREC. Only Temik 15G had improved retention in Zone two, position two when Vydate C-LV<sup>®</sup> treatments were applied at MSU while effects of Aeris<sup>®</sup> at TVREC, were improved over untreated plants when Vydate C-LV<sup>®</sup> was used. Within Zone two, position > 2, all nematicide treatments with Vydate C-LV<sup>®</sup> improved fruit retention compared to untreated plants with the exception of Aeris<sup>®</sup> + Votivo<sup>®</sup> + Vydate C-LV<sup>®</sup> at MSU. Vydate C-LV<sup>®</sup> treatments improved efficacy of the NST especially at the TVREC location.

During bloom evaluation, Zone three (Nodes 15-19) had the greatest difference and benefits from nematicide applications at MSU with Vydate C-LV<sup>®</sup> improving the performance of both Temik 15G and Aeris<sup>®</sup> + Votivo<sup>®</sup> +Vydate C-LV<sup>®</sup> although all treatments were better than the control (Tables 3.14 and 3.15). At TVREC, all treatments resulted in greater fruit retention compared to the untreated plants, but addition of Vydate C-LV<sup>®</sup> did not enhance retention. Fruit retention in Zone three, position two at MSU (Table 3.14) declined in Vydate C-LV<sup>®</sup> treatments with the exception of Aeris<sup>®</sup> + Votivo<sup>®</sup> + Vydate C-LV<sup>®</sup> compared to untreated increased harvest maturity as a result of increased boll retention at position one and two. There was some retention at Zone three, position >2 at TVREC. TVREC possessed the greatest amount of fruit produced within this zone indicating delayed harvest maturity agreeing with previous fruiting development patterns (Jenkins and McCarty, 1995).

During open boll evaluation (Table 3.18 and 3.19), all nematicide treatments enhanced retention at position one within zone one compared to untreated plants regardless of location. Aeris<sup>®</sup> had poorer retention compared to other nematicides regardless of location. Fruit retention at Zone one, position two increased due to nematicidal activities at both locations. Vydate C-LV<sup>®</sup> improved fruit retention with NST's in high *R. reniformis* populations at TVREC. Position > 2 fruit retention was improved with application of Vydate C-LV<sup>®</sup> for NST's at TVREC (Table 3.19) while all nematicides except Aeris<sup>®</sup> improved retention. Plants treated with NST's at MSU did not benefit from the addition of Vydate C-LV<sup>®</sup>.

Within Zone two, position one all nematicide treatments improved retention compared to untreated plants regardless of location. However, Vydate C-LV<sup>®</sup> did not improve retention of this position within Zone two in MSU and only improved Aeris<sup>®</sup> Votivo<sup>®</sup> + Vydate C-LV<sup>®</sup> at TVREC. All nematicide treatments improved fruit retention in Zone two, position two, compared to untreated plants at both locations with exception of Aeris<sup>®</sup> + Votivo<sup>®</sup> and Temik 15G regardless of location. However, at both locations Vydate C-LV<sup>®</sup> increased NST fruit retention resulting in a higher retention than Temik 15G.

Fruit retention at Zone three, position one was improved with Vydate C-LV<sup>®</sup> applications at MSU. Nematicides not receiving foliar applications of Vydate C-LV<sup>®</sup> did not differ from untreated plants. Fruit retention at TVREC in Zone three, position one was improved by foliar applications of Vydate C-LV<sup>®</sup> when compared to the non Vydate C-LV<sup>®</sup> counterpart. At this Zone, NSTs were better or similar in fruit retention to Temik 15G at both locations. At position two, only Aeris<sup>®</sup> and Aeris<sup>®</sup> + Votivo<sup>®</sup> + Vydate C-LV<sup>®</sup> had less fruit retention than untreated at MSU. Only Aeris<sup>®</sup> + Votivo<sup>®</sup> + Vydate C-LV<sup>®</sup> had lower retention compared to untreated or Aeris<sup>®</sup> at TVREC. It is evident that less effective treatments were continuing fruit production and delaying harvest agreeing with findings by Jenkins et al. (1995; 1990a; 1990b).

Data from zones do not track as well as that of positions across zones; however, Vydate C-LV<sup>®</sup> treatments did influence retention at position one within Zones one and two, but were not as dramatic at position two or >2. Fruit retention at position one is very important for yield increase especially in Zones two and three (Jenkins and McCarty, 1995). Therefore, nematicides are important in suppressing *R. reniformis* and improving fruit retention of *G. hirsutum*. Generally, addition of Vydate C-LV<sup>®</sup> did improve efficacy of the NSTs compared to untreated, but data were varied when comparing the same nematicide with or without Vydate C-LV<sup>®</sup>. However, Vydate C-LV<sup>®</sup> does provide an additional option in the overall management of *R. reniformus*.

#### Effects of nematicides on cotton maturity

Nodes above white flower (NAWF), expression of harvest maturity, at both locations indicated NSTs without Vydate C-LV<sup>®</sup> did not differ from untreated plants in maturity (Tables 3.20 and 3.21). Vydate C-LV<sup>®</sup> applications at MSU decreased harvest maturity of the NSTs compared to untreated, but Temik 15G and Aeris<sup>®</sup> + Votivo<sup>®</sup> + Vydate C-LV<sup>®</sup> were similar to Temik 15G. At TVREC, only Aeris<sup>®</sup> + Votivo<sup>®</sup> + Vydate C-LV<sup>®</sup> decreased NAWF compared to the untreated although it was similar to Temik 15G + Vydate C-LV<sup>®</sup> (Mauney 1986). At both locations, NAWF in NSTs with Vydate C-LV<sup>®</sup> applications had a lower NAWF compared to NSTs without Vydate C-LV<sup>®</sup> except Aeris<sup>®</sup> + Vydate C-LV<sup>®</sup> at TVREC. Nodes above white flower trended lower at MSU (Table 3.20) for all treatments because of early season stress.

Node above cracked boll (NACB) conducted during open boll is a later measure of maturity (Jenkins et al., 1996) and showed NSTs at MSU did result in different harvest maturity compared to untreated plants (Table 3.20). Temik 15G was earlier in harvest maturity than NST with Vydate C-LV<sup>®</sup>. All nematicide treatments at MSU (Table 3.20) had earlier harvest maturity when treated with Vydate C-LV<sup>®</sup>. However, at TVREC (Table 3.21), differences in maturity were not as well defined due to early stress and high *R. reniformis* population. At TVREC, Temik 15G + Vydate C-LV<sup>®</sup> had fewer NACB compared to NSTs, but did not differ from NSTs treated with Vydate C-LV<sup>®</sup> or Temik 15G. Evaluation at TVREC occurred prior to the break in maturity for those treatments containing Vydate C-LV<sup>®</sup>, but does indicate Temik 15G promotes earlier cotton maturity. Node of Last Harvestable Boll (NLHB) during open boll showed no differences among nematicides at MSU (Table 3.20), but were slightly higher at the TVREC location in the untreated, Aeris<sup>®</sup> or Temik 15G treatments (Table 3.21).

Harvest maturity was hastened at both locations in all nematicide treatments with Vydate C-LV<sup>®</sup> increasing percentage of open bolls compared to NSTs without Vydate C-LV<sup>®</sup>. Nematode seed treatments had an equal or a higher percentage of open bolls compared to Temik 15G (Tables 3.20 and 3.21). Maturity data can be related to level of fruit retention especially at position one and two. Bolls at these positions provide a very stong sink for photosynthates which reduce growth and promote harvest earliness (Mauney, 1986; Sadras, 1995).

#### Effects of nematicides on monopodial (vegetative) branch and boll formation

Monopodial branch formation occurs after bloom initiation below the node of first fruiting branch, and the degree of monopodial branch formation is often due to row pattern (i.e. skip row vs. solid), plant population and environmental conditions (Mauney, 1986; Jenkins and McCarty, 1995). Monopodial branch number was increased in plants treated with Vydate C-LV<sup>®</sup> compared to untreated. Additionally, Vydate C-LV<sup>®</sup> increased branch number when combined with Aeris<sup>®</sup> or Aeris<sup>®</sup> + Votivo<sup>®</sup> (Table 3.22). Vydate C-LV<sup>®</sup> treatments did not induce monopodial branch production at TVREC nor did it improve monopodial boll numbers produced per branch. However, at MSU, Vydate C-LV<sup>®</sup> treatments did improve monopidial branch production in Aeris<sup>®</sup> and Aeris<sup>®</sup> + Votivo<sup>®</sup> and number of bolls produced when compared with untreated.

#### Effects of nematicide treatments on basal stalk and boll diameter

Basal stalk diameter is important to the cotton plant since massive above-ground biomass must be supported during boll development (Mauney, 1986). All NSTs increased basal stalk diameter compared to untreated plants at MSU (Tables 3.24), but only Temik 15G + Vydate C-LV<sup>®</sup> increased stalk diameter when compared to untreated plants at TVREC (Table 3.25). Addition of Vydate C-LV<sup>®</sup> at MSU only increased the boll diameter of Aeris<sup>®</sup> + Vydate C-LV<sup>®</sup>. At TVREC, Temik 15G treated with foliar applications of Vydate C-LV<sup>®</sup> resulted in greater stalk diameter than untreated plants.

Boll diameter at node nine, below the terminal, except  $Aeris^{\textcircled{W}} + Vydate C-LV^{\textcircled{W}}$  at MSU, was improved in boll development due to nematicides and Vydate  $C-LV^{\textcircled{W}}$  (Table 3.24). Boll diameter at node 12 at MSU was greatest for  $Aeris^{\textcircled{W}}$  or  $Aeris^{\textcircled{W}} + Vydate C-LV^{\textcircled{W}}$  compared to all other treatments and untreated while Temik 15G + Vydate C-LV<sup>(\textcircled{W}</sup> was greatest in TVREC (Tables 3.24 and 3.24). Boll diameter in untreated plants at node 12 at MSU was 20% greater than TVREC. At both locations, Vydate C-LV<sup>(\textcircled{W}</sup> applications increased boll size at node nine compared to nematicide treatments without Vydate C-LV<sup>(\textcircled{W}</sup> except for Aeris<sup>(\textcircled{W})</sup>. In addition, all nematicide treatments increased boll size at node nine compared to NSTs alone at MSU, but at TVREC, under greater nematode populations, neither Aeris<sup>(\textcircled{W})</sup> + Votivo<sup>(\textcircled{W})</sup> nor Aeris<sup>(\textcircled{W})</sup> differed from Temik 15G. Node nine boll diameters at TVREC were larger with Temik 15G + Vydate C-LV<sup>(\textcircled{W})</sup> than NSTs with or without Vydate C-LV<sup>(\textcircled{W})</sup>. Due to early stress at the TVREC location, boll development and size were delayed but had improved boll size when using NSTs or Temik 15G.

At 12 nodes below the terminal, boll diameter at MSU (Table 3.24) was greatest with Aeris<sup>®</sup> + Votivo<sup>®</sup> + Vydate C-LV<sup>®</sup> compared to all other treated or untreated plants. There was no difference between remaining treatments and untreated. However, TVREC (Table 3.25) location did show differences where NSTs increased boll diameter at Node 12 with application of Vydate C-LV<sup>®</sup> except for Aeris<sup>®</sup> + Vydate C-LV<sup>®</sup>. Temik 15G + Vydate C-LV<sup>®</sup> had greater boll diameter compared to other NSTs or untreated. At TVREC, all NSTs improved boll diameter at fruiting position 12, with exception of Aeris<sup>®</sup> + Vydate C-LV<sup>®</sup>.

Differences in boll diameters between nodes nine and 12 below the terminal node indicate progress in boll development between the oldest (node 12) to the youngest (node nine) boll sampled (Tables 3.24 and 3.25). Boll differences at MSU indicate that Temik 15G with or without Vydate C-LV<sup>®</sup> and Aeris<sup>®</sup> + Votivo<sup>®</sup> + Vydate C-LV<sup>®</sup> had similar differences in boll size than other nematicide treatments. In addition, all nematicide treatments had similar differences when compared to untreated plants. TVREC had greater differences in boll diameter btween node nine and 12 because of late maturity due to early season stress and higher populations of *R. reniformis* affecting boll growth. Plants treated with Vydate C-LV<sup>®</sup> had smaller differences in boll diameter when compared to plants that did not receive Vydate C-LV<sup>®</sup>. All nematicide treatments had smaller differences in boll diameter when compared to untreated.

#### Effect of nematicides on cotton yield grown in *R. reniformis* infested soils

Treatment effects upon yield in pounds per acre at both locations showed application of Vydate C-LV<sup>®</sup> increased yields above NSTs alone (Table 3.26). Under lower *R. reniformis* populations at MSU, Aeris<sup>®</sup> + Votivo<sup>®</sup> + Vydate C-LV<sup>®</sup> had greater yield than Aeris<sup>®</sup> + Vydate C-LV<sup>®</sup>. Nematode seed treatments + Vydate C-LV<sup>®</sup> treatments were greater than Temik 15 G without Vydate C-LV<sup>®</sup> but Aeris<sup>®</sup> + Vydate C-LV<sup>®</sup> was lower than Temik 15G + Vydate C-LV<sup>®</sup>. Temik 15G and Aeris<sup>®</sup> + Vydate C-LV<sup>®</sup> did not differ from each other.

Under higher *R. reniformis* populations at TVREC, Aeris<sup>®</sup> + Votivo<sup>®</sup> + Vydate C-LV<sup>®</sup> produced higher yields than Temik 15G and did not differ from Temik 15G + Vydate C-LV<sup>®</sup> as observed at MSU. Aeris<sup>®</sup> + Vydate C-LV<sup>®</sup> yielded lower than Temik 15G + Vydate C-LV<sup>®</sup> at this location indicating its weakness under high *R. reniformis* populations. However, Aeris<sup>®</sup> + Votivo<sup>®</sup> at this location had greater yield than Aeris<sup>®</sup> alone indicating value of the biological nematicide, Votivo<sup>®</sup>, under high *R. reniformis* populations. Conclusively, Vydate C-LV<sup>®</sup> applications improved yield compared to NSTs alone and NSTs alone had higher yield than untreated plants (Wheeler et al., 2014). Under high *R. reniformis* populations, NSTs benefit from Vydate C-LV<sup>®</sup> applications improving fruit retention as has been observed in findings by Lawrence and McLean (2000; 2002; 2003).

## Performance of nematicide treatments under varying populations of *R. reniformis* in greenhouse environments

#### Effect of R. reniformis on root biomass development

In all treatments, with the exception of Temik 15G, as *R. reniformis* population (juvenile and eggs) increased, root mass decreased, which correlated to a reduction in shoot biomass (Table 3.27) (Lawrence and McLean, 1996 a and b). Aeris<sup>®</sup> + Votivo<sup>®</sup> and Aeris<sup>®</sup> had greater root biomass than Temik 15G or untreated plants in absence of nematodes indicating root development suppression by Temik 15G. However, root

biomass was reduced as *R. reniformis* populations increased with the exception of Temik 15G. Under an initial population (*Pi*), *R. reniformis* of 2,500/500 cc of soil, Aeris<sup>®</sup> or Aeris<sup>®</sup> + Votivo<sup>®</sup> had lower root biomass than Temik 15 G, but all had greater root biomass than the untreated. Addition of Votivo<sup>®</sup> to Aeris<sup>®</sup> did improve root biomass compared to Aeris<sup>®</sup> alone. Treatment effects at *Pi* 5,000 and 7,500/500cc, indicated all nematicide treatments had greater root biomass than untreated plants. However, Temik 15G had greater root biomass than NSTs. The NSTs did not differ from each other at *Pi* of 5,000 but Aeris<sup>®</sup>+ Votivo<sup>®</sup> did improve root biomass development at *Pi* of 7,500. As *R. reniformis* numbers increased, root biomass development declined in Aeris<sup>®</sup> and Aeris<sup>®</sup> + Votivo<sup>®</sup> treatments with no decline in Temik 15G treatment. Aeris<sup>®</sup> + Votivo<sup>®</sup> provided better management at higher *R. reniformis* populations than Aeris<sup>®</sup>. However, all treatments for nematode control improved root biomass over untreated plants.

#### Effects of R. reniformis on shoot biomass development

Treatments without *R. reniformis* had greater stem biomass across all nematicide treatments compared to untreated plants (Table 3.27). At *Pi* 2,500, all nematicide treatments had greater shoot biomass than untreated plants with Aeris<sup>®</sup> + Votivo<sup>®</sup> and Temik 15G having greater shoot biomass than Aeris<sup>®</sup>. With Temik 15G at *Pi* 5,000 *R. reniformis* improved shoot biomass development compared to NSTs. Untreated plants had less biomass when compared to plants receiving nematicides. At *Pi* 7,500 Temik 15G and Aeris<sup>®</sup> + Votivo<sup>®</sup> did not differ in shoot biomass production, but all treatments differed from untreated plants. Temik 15G and Aeris<sup>®</sup> + Votivo<sup>®</sup> had greater shoot biomass development than plants treated with Aeris<sup>®</sup>.

#### Egg and juvenile *R. reniformis* populations across nematicide treatments

Juvenile *R. reniformis* populations were similar for untreated plants, plants treated with Temik 15G, or Aeris<sup>®</sup> + Votivo<sup>®</sup> at *Pi* 2500. Both untreated plants and plants treated with Aeris<sup>®</sup> had similar juvenile numbers at *Pi* 5,000. While Temik 15G had fewer juveniles and plants treated with Aeris<sup>®</sup> + Votivo<sup>®</sup> had the most. At *Pi* 7,500, untreated plants and plants treated with Aeris<sup>®</sup> + Votivo<sup>®</sup> had more juveniles compared to plants treated with Temik 15G or Aeris<sup>®</sup>. Temik 15G continued to have the lowest juvenile numbers compared to all other treatments or untreated plants. Nematode populations can be associated with root volume where there is a direct relation between root growth and nematode population development (Lawrence and McLean, 1996 a and b). Temik 15G reduced *R. reniformis* population in greenhouse environments and prevented normal reproduction. Of the NSTs, Aeris<sup>®</sup> + Votivo<sup>®</sup> had greater root mass at *Pi* 7,500 than Aeris<sup>®</sup>, but neither of the NSTs were as effective in managing *R. reniformis* as Temik 15G.

Egg production with Temik 15 G was similar to untreated regardless of Pi, less than Aeris<sup>®</sup> at any Pi, and less than Aeris<sup>®</sup> + Votivo<sup>®</sup> at Pi 2500 and 5,000 (Table 3.27). Temik 15G prevented reproduction, but populations of *R. reniformis* were similar as treated Pi increased. Across NSTs, egg production of *R. reniformis* was greater at Pi2,500 compared to plants treated with Temik 15G or untreated plants. Plants treated with Aeris<sup>®</sup> + Votivo<sup>®</sup> had higher egg numbers at Pi 3000 or 7,500 compared to plants receiving Temik 15G or untreated.

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### Effect of nematicide treatments on cotton growth at varying *R. reniformis* populations under greenhouse environments

Under a controlled greenhouse environment, fruit initiation (NFFB) occurred earlier in plants treated with Temik 15G compared to untreated plants or Aeris<sup>®</sup> with or without Votivo<sup>®</sup> when *Pi* is 0 or 2500. When *Pi* was 5,000 or 7,500, nematicides hastened fruit initiation compared to untreated plants. In absence of *R. reniformis*, Temik 15G delayed NFFB (Table 3.28). Without presence of *R. reniformis*, Phy 375 was able to initiate fruiting at fruiting node six, the genetically controlled NFFB for this variety. The largest differences in NFFB occurred at *Pi* 2,500 where Temik 15G had fruit initiation similar to *Pi* 0. Nematode seed treatments at this population did not differ from untreated plants and initiated fruiting one node higher than Temik 15G. Within *Pi* 5,000 and 7,500, all nematicide treatments fruited at nodes lower than untreated plants, but at these populations did initiate fruiting one node higher at *Pi* of 5,000 and 7,500 for untreated plants. In presence of nematicides, *R. reniformis* at higher *Pi* delayed fruit initiation, however, NFFB remained one node earlier than untreated plants.

Plant height increased across all nematode populations with nematicide treatments compared to untreated plants (Table 3.28). The greatest height reduction occurred in untreated plants at *Pi* 5,000 and 7,500 treatments. In absence of *R. reniformis*, plant height was improved by NSTs. Aeris<sup>®</sup> + Votivo<sup>®</sup>, Aeris<sup>®</sup> and Temik 15G were similar to each other at *Pi* 0 but did show growth advantages compared to untreated. In presence of *R. reniformis* at *Pi* 2,500 treatment, all nematicide treated plants were taller than untreated plants with no difference among NSTs. At *Pi* 5,000, all nematicides improved plant height over untreated plants. Temik 15G and Aeris<sup>®</sup> + Votivo<sup>®</sup> were similar while

Aeris<sup>®</sup> plants were shorter than other nematicide treated plants. All nematicide treated plants were taller at *Pi* 7,500 than untreated plants. At this population, Temik 15G treated plants had greater plant height than NSTs but NSTs were still taller than untreated plants. Temik 15G offered greater management of *R. reniformis* across a greater nematode population than NSTs. This indicates a need for additional pesticide treatments, i.e. Vydate C-LV<sup>®</sup>, to maintain *G. hirsutum* growth under high populations of *R. reniformis* when using NSTs.

*Rotylenchulus reniformis* affected nodal development and effects on total node (TN) with plants receiving Aeris<sup>®</sup> + Vydate C-LV<sup>®</sup> having more total nodes than other nematicides through *Pi* 5,000 (Table 3.28). Height node ratios were similar for treated or untreated plants at *Pi* 0. However, nematicides increased HNR compared to untreated plants at p 2500 or 5,000. All NSTs increased TN across all *R. reniformis* populations. All nematicides in absence of *R. reniformis* improved TN development compared to untreated plants indicating nematicides enhanced *G. hirsutum* growth. Height to node ratio at *Pi* 7,500, showed plants treated with Aeris<sup>®</sup> + Votivo<sup>®</sup> continued to produce more nodes than Aeris<sup>®</sup> even with a greater HNR. While Temik 15G plants had fewer nodes, HNR was greater under increasing *R. reniformis* populations than Aeris<sup>®</sup> or Aeris<sup>®</sup> + Votivo<sup>®</sup>.

#### Conclusion

*R. reniformis* greatly affected all growth aspects of *G. hirsutum*, including NFFB, plant height, boll size, internode elongation, fruit retention during bloom and open boll growth phases at fruiting positions one and two, delayed maturity and reduced yield. However, use of nematicides improved *G. hirsutum* performance in *R. reniformis* infested soils similar to Phipps and Eisenback (2005). Temik 15G was generally the best standalone nematicide treatment as observed in field and greenhouse studies. Of the NSTs, Aeris<sup>®</sup> + Votivo<sup>®</sup> provided best growth under *R. reniformis* infested soils. Nematicide seed treatments alone improved performance of *G. hirsutum* compared to untreated plants in most growth parameters. However, efficacy of NSTs was usually improved with foliar applications of Vydate C-LV<sup>®</sup> making them comperable to Temik 15G without Vydate C-LV<sup>®</sup>. Temik 15G efficacy was generally improved with Vydate C-LV<sup>®</sup> applications, but Vydate C-LV<sup>®</sup> tended to not impact efficacy of Temik 15G as much as in NSTs. Under high *R. reniformis* populations, NSTs alone did not offer satisfactory management of *R. reniformis* without additional control from Vydate C-LV<sup>®</sup>.

Treatment	Rate	Mode of Application
Aeris <sup>®</sup> + Orthene 90 S	.075 mg ai/seed rate + 0.75 Lbs ai/Ac	Seed treatment followed by foliar applications at 6 leaf and 10 leaf
Aeris <sup>®</sup> + Votivo <sup>®</sup> + Orthene 90 S Temik 15G <sup>y</sup> + Orthene 90 S	0.424 mg ai/seed rate+ 0.75 Lbs ai/Ac 0.75 lbs ai/ac + 0.75 Lbs ai/Ac	Seed reatment followed by foliar applications at 6 leaf and 10 leaf At-planting followed by foliar applications at 6 leaf and 10 leaf
Aeris <sup>®</sup> +Vydate C-LV <sup>®</sup> x + Vydate C-LV <sup>®</sup>	0.075 mg ai/seed rate; + 8.0 Oz/ac; + 8.0 Oz/ac	Seed Treatment followed by foliar applications at 6 leaf and 10 leaf
Aeris <sup>®</sup> + Votivo <sup>®</sup> + Vydate C-LV <sup>®</sup> +Vydate C-LV <sup>®</sup>	0.075 mg ai/seed rate; + 0.424 mg ai/seed rate; + 0.24 Lbs ai/ac; + 0.24 Lbs ai/ac	Seed treatment followed by foliar applications at 6 leaf and 10 leaf
Temik 15G + Vydate C-LV <sup>®</sup> + Vydate C-LV <sup>®</sup>	0.75 lbs ai/ac; + 0.24 Lbs ai/ac; + 0.24 Lbs ai/ac	At-planting followed by foliar applications at 6 leaf and 10 leaf

In- the- field treatment list for seed applied nematicides (Aeris<sup>®</sup> and Aeris<sup>®</sup> Table 3.1 + Votivo<sup>®</sup>), at-planting hopper box treatment (Temik 15G) and in-season foliar application (Vydate C-LV<sup>®</sup>) applied with CO<sub>2</sub> back-pack sprayer.

<sup>z</sup> Aeris<sup>®</sup> and Aeris<sup>®</sup> + Votivo<sup>®</sup> were applied to the seed prior to planting by Bayer Crop Science at Research Triangle Park, Raleigh, North Carolina.

<sup>y</sup> Temik 15G was applied at planting via hopper boxes pre-calibrated to apply the product in-furrow beneath the seed.

<sup>x</sup> Vydate C-LV<sup>®</sup> was applied to the foliage using a CO<sub>2</sub> backpack sprayer calibrated to deliver 15.0 gallons of water per acre.

Treatment	Rate	Mode of Application	Innoculated reniform numbers
Aeris®	0.075 mg ai/seed rate	Seed Treatment	0
Aeris <sup>®</sup> + Votivo <sup>®</sup>	0.075 mg ai/seed rate + 0.1424 mg ai/seed rate	Seed Treatment	0
Temik 15G	0.75 lbs ai/ac	At-Planting	0
Untreated	-	-	0
Aeris®	0.075 mg Ai/seed rate	Seed Treatment	2,500
Aeris <sup>®</sup> + Votivo <sup>®</sup>	.075 mg ai/seed rate + 0.424 mg ai/seed rate	Seed Treatment	2,500
Temik 15G	0.75 lbs ai/ac	At-Planting	2,500
Untreated	-	-	2,500
Aeris®	0.075 mg ai/seed rate	Seed Treatment	5,000
Aeris <sup>®</sup> + Votivo <sup>®</sup>	.075 mg ai/seed rate + 0.424 mg ai/seed rate	Seed Treatment	5,000
Untreated	-	-	5,000
Aeris®	0.075 mg ai/seed rate	Seed Treatment	7,500
Aeris <sup>®</sup> + Votivo <sup>®</sup>	0.075 mg ai/seed rate + 0.424 mg ai/seed rate	Seed Treatment	7,500
Temik 15G	0.75 lbs ai/ac	At-Planting	7,500

Table 3.2Treatment list for greenhouse nematicide study where Phy 375 was grown<br/>under varying *R. reniformis* populations in a autoclaved pre-mixed soil.

Treatment		Reniform	Nematode Numbe	ers/500 cc
	May	Square	Bloom	Open Boll
	$(0 \text{ DAE}^{x})$	(40 DAE)	(70 DAE)	(100 DAE)
Untreated	4726.0a <sup>z</sup>	2761.0a	4982.0d	8973.6c
Temik 15G	4541.0a	928.0d	9306.0a	16092.8b
Aeris®	5308.0a	1821.0b	5828.6cd	15263.6b
Aeris <sup>®</sup> + Votivo <sup>®</sup>	4107.0a	1551.0bc	6181.0cd	16622.8b
Temik 15G + Vydate	_ y	578.00e	9490.6a	23026.6a
C-LV®				
Aeris <sup>®</sup> + Vydate C-	-	1418.0c	7922.8ab	22704.0a
$LV^{\mathbb{R}}$				
Aeris <sup>®</sup> + Votivo <sup>®</sup> +	-	1031.0d	7236.6bc	18105.4b
Vydate C-LV <sup>®</sup>				
LSD (0.05)	3381.0	307.6	1297.4	2990.9

Table 3.3Seasonal progression of *R. reniformis* sampled at six core samples per plot<br/>during four growth stages at Mississippi State University.

<sup>y</sup> Indicates no data taken at this evaluation since Vydate C-LV<sup>®</sup> had not been applied.

<sup>x</sup> DAE=Days after emergence.

Table 3.4Seasonal progression of *R. reniformis* sampled at six core samples per plot<br/>during four growth stages at Tennessee Valley Research and Extension<br/>Center.

Treatment		Reniform	Nematode Numbe	ers/500 cc
	May	Square	Bloom	Open Boll
	$(0 \text{ DAE}^{x})$	(40 DAE)	(70 DAE)	(100 DAE)
Untreated	22252.5b <sup>z</sup>	21901.3a	5848.0c	7625.3d
Temik 15G	27755.9a	6536.0cd	9030.0bc	16015.1bc
Aeris®	21376.4b	11008.0b	8428.0bc	11829.5c
Aeris <sup>®</sup> + Votivo <sup>®</sup>	20981.9b	11180.0b	8886.7bc	13416.0c
Temik 15G + Vydate	_ y	5188.8d	13588.0a	24710.7a
C-LV®				
Aeris <sup>®</sup> + Vydate C-	-	9508.3bc	9173.3bc	16301.7bc
LV®				
Aeris <sup>®</sup> + Votivo <sup>®</sup> +	-	7138.0cd	12040.0b	20668.7b
Vydate C-LV <sup>®</sup>				
LSD (0.05)	1549.1	2628.5	2898.4	67.8

<sup>z</sup> Means within columns followed by the same letter are not different according to the Least Significant Difference means separation test  $P\alpha$ =0.05.

<sup>y</sup> Indicates no data taken at this evaluation since Vydate C-LV<sup>®</sup> had not been applied.

<sup>x</sup> DAE=Days after emergence.

Table 3.5Plants/ac (in 1000's), visual vigor and hypocotyl lengths acquired 14 days<br/>after emergence at Mississippi State University and Tennessee Valley<br/>Research and Extension Center.

Treatment	_	MS	U	TVREC				
	Plants/ac	Vigor	Hypocotyl	Plants/ac	Vigor	Hypocotyl		
	(1000's) <sup>w</sup>	$(1-5)^{x}$	(mm) <sup>y</sup>	(1000's)	(1-5)	(mm)		
Untreated	51.7a <sup>z</sup>	2.1a	9.6b	33.1a	3.3a	6.7d		
Temik 15G	49.9c	1.0c	10.8a	27.2ab	1.9c	7.1c		
Aeris®	49.1b	1.5b	10.4a	29.8ab	2.4b	7.2bc		
Aeris <sup>®</sup> +	50.2c	1.5b	10.8a	24.8b	1.5c	7.5b		
Votivo®								
LSD (0.05)	1.0	0.4	0.3	1.0	0.3	0.1		

<sup>y</sup> Hypocotyl measured from point of seed attachment to cotyledon.

<sup>x</sup> Visual vigor evaluation in ranking of 1-5 where 1 had best growth and 5 the lowest.

<sup>w</sup> Plants/ac was conducted by counting all plants per plot with expanded cotyledons.

Table 3.6Node of first fruiting branch (NFFB) acquired during square (40 days after<br/>emergence) at Mississippi State University and Tennessee Valley Research<br/>and Extension Center.

Treatment	NF	FFB
	MSU	TVREC
Untreated	7.0a <sup>z</sup>	7.5a
Temik 15G	5.8c	6.6b
Aeris®	6.3b	7.2ab
Aeris <sup>®</sup> +	6.3b	6.9b
Votivo®		
LSD (0.05)	0.5	0.7

Z Means within columns followed by the same letter are not different according to the Least Significant Difference means separation test Pα=0.05.

Treatment		Square	x,y		Blo	om		Open	Boll
-	PH	TN	HNR	PH	TN	HNR	PH	TN	HNR
Untreated	12.8dc <sup>z</sup>	12.2d	1.2c	31.3b	17.9d	1.7a	37.4f	23.2bc	1.6c
Temik 15G	15.1abc	12.8bc	1.0d	33.1a	18.5c	1.8a	39.0d	21.3bc	1.8b
Aeris®	14.7c	12.4d	1.1c	31.8b	18.3c	1.8a	38.1e	21.2a	1.8b
Aeris <sup>®</sup> +	14.9bc	12.7c	1.3a	32.5ab	18.5c	1.8a	38.9d	21.6bc	1.8b
Votivo®									
Temik 15G +	15.8a	13.3a	1.2b	34.3a	18.8bc	1.8a	40.9a	20.5c	1.99a
Vydate C-									
LV®									
Aeris <sup>®</sup> +	15.5ab	13.4a	1.1c	33.1a	19.1b	1.7a	39.5c	21.9b	1.8b
Vydate C-									
LV®									
Aeris <sup>®</sup> +	15.8a	13.2a	1.2b	33.3a	19.5a	1.8a	40.2b	20.6bc	1.93a
Votivo <sup>®</sup> +									
Vydate C-									
LV®									
LSD (0.05)	0.7	0.3	0.1	1.6	0.5	0.1	0.6	0.8	0.1

Table 3.7Cotton growth parameters, plant height (inches), total nodes and height to<br/>node ratio (inches), at square, bloom and open boll stages in *R. reniformis*<br/>infested soils at Mississippi State University.

<sup>y</sup> Evaluation timing Days After Emergence (DAE); square (40 DAE); bloom (70 DAE); open boll (100 DAE).

<sup>x</sup> Average six consecutive plants with normal terminal per plot sampled destructively.

Treatment Bloom Open Boll Square x,y PH ΤN HNR PH TN HNR PH TN HNR Untreated 11.0b 0.5d 15.8d 18.0d 0.9e 23.4d 26.0a 0.9d 5.5d<sup>z</sup> Temik 15G 8.7b 13.0a 0.7b 23.4b 19.0bc 1.2b 28.5bc 24.0c 1.2bc Aeris® 5.7d 20.4c 19.0c 1.0cd 24.0c 11.0b 0.5d 27.5c 1.1c Aeris<sup>®</sup> + 7.1c 20.5c 19.0bc 1.0d 28.0bc 24.0c 1.2bc 12.0b 0.6c Votivo® Temik 15G + 9.9a 12.0a 0.8a 26.6a 20.0a 1.3a 29.8b 25.0c 1.2b Vydate C-LV® Aeris ®+ 8.4b 11.0b 0.7b 22.0bc 19.0bc 1.1bc 32.7a 25.0b 1.3a Vydate C-LV® Aeris® + 8.9b 13.0a 0.7b 23.2b 20.0ab 28.8b 25.0b 1.2bc 1.2b Votivo<sup>®</sup> + Vydate C-LV® LSD (0.05) 0.6 0.7 0.1 1.7 0.7 0.9 1.6 0.7 0.1

Table 3.8Cotton growth parameters, plant height (inches), total nodes and height to<br/>node ratio (inches), at square, bloom and open boll stages in *R. reniformis*<br/>infested soils across time at Tennessee Valley Research Extension Center.

<sup>y</sup> Evaluation timing Days After Emergence (DAE); square (40 DAE); bloom (70 DAE); open boll (100 DAE).

<sup>x</sup> Average six consecutive plants with normal terminal per plot sampled destructively.

Table 3.9	Average plant height (inches) at each node culminating in total height
	(inches) of cotton measured during square (40 days after emergence) in R.
	reniformis infested soils at Mississippi State University.

				Plant heigl	nt at each no	de (inches	<u>)</u> x			
Treatment	Node Number <sup>y</sup>									
_	1	3	5	7	9	11	13	15		
Untreated	1.5b <sup>z</sup>	3.4b	6.3a	9.0a	11.7a	12.9a	13.2b	13.3b		
Temik 15G	1.8a	4.3a	6.4a	9.0a	12.1a	14.0a	14.5ab	14.6ab		
Aeris®	1.6a	4.3a	6.2a	8.6a	11.5a	13.4a	14.0ab	14.0ab		
Aeris <sup>®</sup> + Votivo <sup>®</sup>	1.6a	4.2a	6.2a	8.8a	11.7a	13.5a	14.1ab	14.2ab		
Temik <sup>®</sup> + Vydate C-LV <sup>®</sup>	1.7a	4.5a	6.4a	9.5a	12.2a	14.5a	15.5a	15.5a		
Aeris <sup>®</sup> + Vydate C-LV <sup>®</sup>	1.7a	4.6a	6.6a	9.2a	12.1a	13.7a	14.2ab	14.3ab		
Aeris <sup>®</sup> + Votivo <sup>®</sup> + Vydate C-LV <sup>®</sup>	1.8a	4.5a	6.4a	9.2a	12.3a	14.3a	14.9ab	15.0ab		
LSD (0.05)	0.2	0.6	0.8	1.0	1.2	1.2	1.3	1.4		

<sup>z</sup> Means within columns followed by the same letter are not different according to the Least Significant Difference means separation test  $P\alpha$ =0.05.

<sup>y</sup> Odd node measurements are shown to facilitate reporting.

<sup>x</sup> Average six consecutive plants with normal terminal per plot sampled destructively.

Table 3.10Average plant height (inches) at each node culminating in total height<br/>(inches) of cotton measured during square (40 days after emergence) in R.<br/>reniformis infested soils at Tennessee Valley Research Extension Center.

Treatment		Plant height at each node (inches) <sup>x</sup>									
				Node	Number <sup>y</sup>						
-	1	3	5	7	9	11	13	15			
Untreated	0.87cd <sup>z</sup>	1.7c	2.6d	3.6e	4.8d	5.3e	5.4f	5.4f			
Temik 15G	0.84cd	1.8c	2.8d	4.2d	5.1c	6.2d	6.8d	6.9d			
Aeris®	0.83cd	1.8c	2.7d	3.8e	4.8d	5.3e	5.4f	5.4f			
Aeris <sup>®</sup> + Votivo <sup>®</sup>	0.77d	1.9c	2.6d	3.7e	5.2c	6.3d	6.5e	6.5e			
Temik 15 G + Vydate C-LV <sup>®</sup>	1.08a	2.9a	4.1a	5.7a	7.4a	8.9a	9.6a	9.7a			
Aeris <sup>®</sup> + Vydate C-LV <sup>®</sup>	1.0b	2.4b	3.8b	5.4b	7.1b	8.2c	8.6c	8.6c			
Aeris <sup>®</sup> + Votivo <sup>®</sup> + Vydate C-LV <sup>®</sup>	1.02b	2.3b	3.4c	5.1c	7.5a	8.5b	8.8b	8.8b			
LSD (0.05)	0.1	0.1	0.2	0.3	0.2	0.2	0.1	0.1			

<sup>y</sup> Odd node measurements are shown to facilitate reporting.

<sup>x</sup> Average six consecutive plants with normal terminal per plot sampled destructively.

Table 3.11Average plant height (inches) at each node culminating in total height<br/>(inches) of cotton measured during bloom (70 days after emergence) in R.<br/>reniformis infested soils at Tennessee Valley Research Extension Center.

Treatment				Pla	nt heigh	t at each	node (in	hes) <sup>x</sup>			
		Node Number <sup>y</sup>									
	1	3	5	7	9	11	13	15	17	19	21
Untreated	0.85b <sup>z</sup>	1.7c	2.4d	3.6d	5.4e	8.5e	11.4e	14.3f	15.6f	16.4f	16.6f
Temik 15G	1.0a	2.5a	4.0a	5.3b	7.4b	11.1b	15.5b	20.9c	24.6a	26.4b	26.6b
Aeris®	1.02a	2.3b	3.5c	4.6c	6.1d	8.6e	12.4d	16.8e	19.3f	20.0e	20.2e
Aeris®+	1.03a	2.6a	3.7b	5.3b	7.2bc	9.1d	12.6d	17.1e	20.8e	22.7d	23.1d
Votivo®											
Temik 15G +	1.04a	2.6a	3.7b	5.7a	8.3a	11.3ab	16.4a	22.7a	25.7b	27.2a	27.8a
Vydate C-LV®											
Aeris <sup>®</sup> +	1.06a	2.6a	3.7b	4.7c	6.9c	10.3c	14.8c	19.8d	22.5c	23.3c	23.3c
Vydate C-LV®											
Aeris <sup>®</sup> +	1.08a	2.6a	3.8ab	5.7a	8.0a	11.5a	16.5a	21.6b	23.2c	23.7c	23.9c
Votivo <sup>®</sup> +											
Vydate C-LV®											
LSD (0.05)	0.14	0.2	0.2	0.26	0.3	0.4	0.6	0.5	0.6	0.7	0.7

<sup>z</sup> Means within columns followed by the same letter are not different according to the Least Significant Difference means separation test  $P\alpha=0.05$ .

<sup>y</sup> Odd node measurements are shown to facilitate reporting.

<sup>x</sup> Average six consecutive plants with normal terminal per plot sampled destructively.

Treatment	%	Retentior	nw	%	6 Retentio	n	% Retention		
	(40 DAE) <sup>v</sup>				(70 DAE)		(100 DAE)		
	Pos 1 <sup>y</sup>	Pos 2	Pos >2	Pos <sup>y</sup> 1	Pos 2	Pos >2	Pos 1	Pos 2	Pos >2
Untreated	99.2a <sup>z</sup>	72.6d	26.2e	76.3d	36.1b	17.7b	49.4d	19.6d	3.9bc
Temik 15G	99.4a	75.1bc	50.1b	85.6c	52.4a	30.0b	66.3a	23.9c	2.9c
Aeris®	100.0a	74.1bc	32.7d	83.3c	40.1b	43.6a	54.8c	24.4c	5.2b
Aeris <sup>®</sup> + Votivo <sup>®</sup>	99.4a	76.7ab	45.1c	86.6bc	37.4b	51.5a	59.8b	26.4c	7.4a
Temik 15G +	100.0a	79.2a	53.4a	94.4a	53.9a	25.4b	68.8a	33.8a	0.0d
Vydate C-LV <sup>®</sup>									
Aeris <sup>®</sup> + Vydate	99.0a	78.8ab	53.4a	84.3c	51.4a	25.7b	61.5b	29.9b	7.1a
C-LV®									
Aeris <sup>®</sup> + Votivo <sup>®</sup>	99.2a	76.1ab	50.4ab	89.2b	52.5a	25.1b	68.6a	30.4b	2.8c
+ Vydate C-LV®									
LSD (0.05)	1.8	3.0	3.6	3.3	7.3	10.8	3.3	2.6	1.9

Table 3.12Percent (%) fruit retention at sympodial positions 1, 2 and > 2 during<br/>square, bloom and open boll stages at Mississippi State University.

<sup>y</sup> Pos=Sympodial (fruiting) position.

 $\times$  % retention was by fruiting position across the whole plant.

<sup>w</sup> Average six plants with normal terminal sampled destructively per plot.

<sup>v</sup> Evaluation timings (Days After Emergence); square-June (40 DAE); bloom-July (70 DAE); open boll-Sept. (100 DAE).

Treatment	% Retention <sup>w</sup>			% Retention			% Retention		
	$(40 \text{ DAE})^{\text{V}}$			(70 DAE)			(100 DAE)		
	Pos 1 <sup>y</sup>	Pos 2	Pos >2	Pos 1	Pos 2	Pos >2	Pos 1	Pos 2	Pos >2
Untreated	98.3a <sup>z</sup>	13.5d	0.0a	70.5b	27.2d	1.4c	45.9c	16.8d	3.8c
Temik 15G	98.3a	50.9a	0.0a	95.9a	71.2a	65.5ab	51.3b	20.8c	7.9b
Aeris®	98.6a	21.2cd	0.0a	95.2a	46.2c	35.2sbc	46.8c	22.2c	10.2b
Aeris <sup>®</sup> + Votivo <sup>®</sup>	100.0a	24.6c	0.0a	93.9a	58.0b	24.3bc	47.5c	17.4d	9.7b
Temik 15G +	100.0a	50.9a	0.0a	96.9a	70.6a	76.9a	53.8a	28.0a	14.1a
Vydate C-LV <sup>®</sup>									
Aeris <sup>®</sup> + Vydate	100.0a	33.5b	0.0a	94.1a	53.7b	36.2abc	53.2a	25.5b	15.3a
C-LV®									
Aeris <sup>®</sup> + Votivo <sup>®</sup>	97.5a	47.2a	0.0a	94.1a	65.9a	41.1abc	55.1a	29.2a	10.8b
+ Vydate C-LV®									
LSD (0.05)	2.9	10.6	0.0	5.4	6.5	35.5	1.9	2.1	3.6

Table 3.13Percent (%) fruit retention at sympodial positions 1, 2 and > 2 during<br/>square, bloom and open boll stages at Tennessee Valley Research Extension<br/>Center.

<sup>y</sup> Pos=Sympodial (fruiting) position.

x % retention was by fruiting position across the whole plant.

<sup>w</sup> Average six plants with normal terminal sampled destructively per plot.

<sup>v</sup> Evaluation timings Days After Emergence (DAE); square-June (40 DAE); bloom-July (70 DAE); open boll-Sept. (100 DAE).

Table 3.14Percent (%) fruit retention by sympodial positions 1, 2 and >2 within zons<br/>as divided by nodes during square (40 days after emergence) at Mississippi<br/>State University.

Treatment	Zor	ne 1 (Nodes 5-9	<u>)</u> x	Zone 2 (Nodes 10-14)				
		(%)		(%)				
	Pos 1 <sup>y</sup>	Pos 2	Pos>2	Pos 1	Pos 2	Pos >2		
Untreated	98.0a <sup>z</sup>	91.4b	50.4b	100.0a	51.8b	0.33a		
Temik 15G	99.6a	97.0ab	87.1a	100.0a	59.1ab	8.3a		
Aeris®	98.4a	93.5ab	55.9b	100.0a	53.0ab	2.1a		
Aeris <sup>®</sup> +	98.4a	92.1b	51.3b	100.0a	53.1ab	0.78a		
Votivo®								
Temik 15G +	100.0a	99.2a	96.9a	100.0a	66.2a	3.3a		
Vydate C-LV®								
Aeris <sup>®</sup> +	98.8a	99.2a	95.0a	100.0a	61.7ab	1.6a		
Vydate C-LV®								
Aeris <sup>®</sup> +	100.0a	96.5ab	98.4a	100.0a	58.5ab	3.1a		
Votivo <sup>®</sup> +								
Vydate C-LV®								
LSD (0.050)	3.2	4.8	12.3	1.4	8.7	6.2		

<sup>y</sup> Pos=Sympodial (fruiting) Position.

<sup>x</sup> Zone 1 represents fruit retention between nodes 5 to 9; Zone 2 represents fruit retention between nodes 10 to 14.

<sup>w</sup> Average six plants with normal terminal sampled destructively per plot.

Table 3.15Percent (%) fruit retention<sup>v</sup> by sympodial positions 1, 2 and >2 within<br/>zones as divided by nodes during square (40 days after emergence) at<br/>Tennessee Valley Research Extension Center.

Treatment	Zoi	ne 1 (Nodes 5-9 (%)	9) <sup>w</sup>	Zone 2 (Nodes 10-14) (%)			
	Pos 1 <sup>x</sup>	Pos 2	Pos>2	Pos 1	Pos 2	Pos>2	
Untreated	96.3a <sup>z</sup>	22.9c	_ y	75.0b	0.0c	-	
Temik 15G	99.4a	84.6a	-	100.0a	2.6c	-	
Aeris®	100.0a	34.4c	-	94.9a	5.6bc	-	
Aeris <sup>®</sup> +	100.0a	49.3bc	-	97.2a	4.2bc	-	
Votivo®							
Temik 15G +	100.0a	84.2a	-	97.2a	19.6a	-	
Vydate C-LV <sup>®</sup>							
Aeris <sup>®</sup> +	100.0a	64.6ab	-	100.0a	12.5abc	-	
Vydate C-LV <sup>®</sup>							
Aeris <sup>®</sup> +	100.0a	88.9a	-	100.0a	17.4ab	-	
Votivo <sup>®</sup> +							
Vydate C-LV <sup>®</sup>							
LSD (0.050)	4.2	25.5	0.0	28.5	11.7	0.0	

<sup>y</sup> (-) reflects no data at fruiting position >2.

<sup>x</sup> Pos=Sympodial (Fruiting) Position.

<sup>w</sup> Zone 1 represents fruit retention between nodes 5 to 9; Zone 2 represents fruit retention between nodes 10 to 14.

<sup>v</sup> Average six plants with normal terminal sampled destructively per plot.

Table 3.16Percent (%) fruit retention<sup>v</sup> by sympodial positions 1, 2 and >2 within zones<br/>as divided by nodes during bloom (70 days after emergence) at Mississippi<br/>State University.

Treatment	Zone 1 (Nodes 5-9) <sup>W</sup>			Zone	Zone 2 (Nodes 10-14)			Zones 3 (Nodes 15-19)		
	(%)				(%)			(%)		
-	Pos 1 <sup>x</sup>	Pos 2	Pos>2	Pos 1	Pos 2	Pos>2	Pos 1	Pos 2	Pos >2	
Untreated	67.8c <sup>z</sup>	37.1b	36.1b	86.7b	32.8c	21.7c	60.0d	8.3d	_y	
Temik 15G	75.8abc	61.7a	33.8b	98.9a	72.5b	36.1bc	83.8bc	25.0ab	-	
Aeris®	69.0c	40.1b	50.1b	93.3a	65.0b	26.7bc	83.3bc	30.6a	-	
Aeris <sup>®</sup> +	78.3abc	45.1b	43.7b	100.0a	61.7b	32.5bc	81.7c	26.7ab	-	
Votivo®										
Temik 15G +	85.5a	62.6a	97.9a	100.0a	86.7a	41.6b	97.8a	13.3cd	-	
Vydate C-LV <sup>®</sup>										
Aeris <sup>®</sup> + Vydate	78.6abc	69.0a	50.3b	100.0a	68.3b	56.7a	91.5ab	18.3bc	-	
C-LV®										
Aeris <sup>®</sup> +	84.2ab	67.2a	99.1a	100.0a	66.7b	31.6bc	93.3a	12.5cd	-	
Votivo <sup>®</sup> +										
Vydate C-LV <sup>®</sup>										
LSD (0.05)	10.8	16.0	21.9	4.0	12.2	15.3	8.3	8.2	0.0	

<sup>y</sup> (-) reflects no data at fruiting position  $\geq 2$ .

<sup>x</sup> Pos=Sympodial (Fruiting) Position.

<sup>w</sup> Zone 1 represents fruit retention between nodes 5 to 9; Zone 2 represents fruit retention between nodes 10 to 14; Zone 3 represent fruit retained between nodes 15 to 19.

<sup>v</sup> Average six plants with normal terminal sampled destructively per plot.

Treatment Zone 2 (Nodes 10-14) Zones 3 (Nodes 15-19) Zone 1 (Nodes 5-9)<sup>x</sup> (%) (%) (%) Pos 2 Pos > 2Pos 1 Pos 2 Pos > 2Pos 1 Pos 2 Pos > 2Pos 1<sup>y</sup> 70.8b<sup>z</sup> 53.9c 21.7c 0.0b 5.6c 4.2c 86.7b 54.4c 0.0f Untreated Temik 15G 92.6a 59.1a 40.8b 100.0a 90.0ab 97.0a 95.0ab 63.3a 3.3b Aeris® 97.2a 39.6b 38.2b 100.0a 60.0c 44.3d 88.3b 43.3b 0.0b Aeris® + 91.2a 35.1b 50.7ab 96.4a 78.3b 61.8c 95.0ab 43.3b 9.9a Vydate C-LV® Temik 15 G + 90.8a 61.8a 57.9a 100.0a 95.0a 100.0a 100.0a 60.0b 1.3b Vydate C-LV® Aeris® + 85.0a 30.6b 35.0b 98.3a 90.0ab 39.6e 87.5b 44.4b 0.0b Votivo® Aeris® + 94.7a 63.5a 49.1ab 100.0a 91.1ab 76.3b 98.3ab 48.3b 0.0 Votivo<sup>®</sup> + Vvdate C-LV® LSD (0.05) 11.0 8.9 14.2 6.3 5.1 4.3 5.7 7.3 0.0

Table 3.17 Percent (%) fruit retention<sup>w</sup> by sympodial positions 1, 2 and >2 within zones as divided by nodes during bloom (70 days after emergence) at Tennessee Valley Research Extension Center.

<sup>y</sup> Pos=Sympodial (Fruiting) Position.

<sup>x</sup> Zone 1 represents fruit retention between nodes 5 to 9; Zone 2 represents fruit retentionbetweennodes 10 to 14; Zone 3 represent fruit retained between nodes 15 to 19.

<sup>w</sup> Average six plants with normal terminal sampled destructively per plot.

Table 3.18Percent (%) fruit retentionv by sympodial positions 1, 2 and >2 within zones<br/>as divided by nodes during open boll (100 days after emergence) at<br/>Mississippi State University.

Treatment	Zone 1 (Nodes 5-9) $^{\text{w}}$ (%)		Zone 2 (Nodes 10-14) (%)			Zones 3 (Nodes 15-19) (%)			
	Pos 1 <sup>x</sup>	Pos 2	Pos >2	Pos 1	Pos 2	Pos >2	Pos 1	Pos 2	Pos >2
Untreated	31.7c <sup>z</sup>	25.5c	5.0c	73.3c	23.3c	6.7a	40.0b	10.0a	_у
Temik 15G	58.3a	41.7b	8.9bc	81.6ab	23.3c	0.0c	43.3b	6.7ab	-
Aeris®	49.4b	39.4b	16.3a	81.8ab	31.6b	6.7a	41.6b	0.0c	-
Aeris <sup>®</sup> +	56.1a	38.1b	8.8bc	81.6ab	25.0c	8.9a	41.6b	6.7ab	-
Votivo®									
Temik 15G +	61.3a	42.2b	0.0d	83.9a	41.6a	0.0c	63.7a	6.2ab	-
Vydate C-LV®									
Aeris <sup>®</sup> +	56.7a	43.3b	13.3ab	80.0b	41.3a	5.0b	62.2a	6.8ab	-
Vydate C-LV®									
Aeris <sup>®</sup> +	58.9a	51.6a	8.3bc	83.3ab	33.9b	0.0c	62.7a	2.5bc	-
Votivo <sup>®</sup> +									
Vydate C-LV®									
LSD (0.05)	6.8	5.3	4.5	2.9	4.6	3.1	5.8	4.2	0.0

<sup>z</sup> Means within columns followed by same letter are not different according to Least Significant Difference means separation test  $P\alpha$ =0.05.

<sup>y</sup> (-) reflects no data at fruiting position >2.

<sup>x</sup> Pos=Sympodial (Fruiting) Position.

<sup>w</sup> Zone 1 retention nodes 5 to 9; Zone 2 retention nodes 10 to 14; Zone 3 retention nodes 15 to 19.

<sup>v</sup> Average six plants with normal terminal sampled destructively per plot.

Treatment	Zone 1 (Nodes 5-9) <sup>x</sup>		Zone 2 (Nodes 10-14)		Zones 3 (Nodes 15-19)				
	(%)			(%)		(%)			
	Pos 1 <sup>y</sup>	Pos 2	Pos >2	Pos 1	Pos 2	Pos >2	Pos 1	Pos 2	Pos >2
Untreated	18.3d <sup>z</sup>	5.0d	0.0d	73.5c	35.6de	10.0b	58.3e	22.2ab	5.0a
Temik 15G	52.3b	35.0a	13.9bc	81.9b	31.7e	17.8ab	68.3bc	16.9abc	0.0b
Aeris®	33.3c	23.8b	6.9d	79.4b	45.0c	28.3a	60.9de	23.3a	5.0a
Aeris <sup>®</sup> +	46.7b	11.1c	15.0c	85.0b	39.4d	28.3a	64.4cd	15.0abc	0.0b
Votivo <sup>®</sup>									
Temik 15G +	53.3b	35.6a	25.0ab	86.1b	49.4b	26.5a	78.3a	13.3bc	5.0a
Vydate C-LV <sup>®</sup>									
Aeris <sup>®</sup> +Vydate	47.5b	25.6b	31.1a	86.1b	68.3a	25.0	68.3bc	18.3abc	5.0a
C-LV <sup>®</sup>									
Aeris <sup>®</sup> +	63.3a	38.3a	18.3b	92.8a	66.7a	15.6ab	73.2b	11.7c	5.0a
Votivo <sup>®</sup> +									
Vydate C-LV <sup>®</sup>									
LSD (0.05)	7.6	4.8	7.8	9.4	9.8	9.7	5.7	7.3	0.0

Table 3.19Percent (%) fruit retention\* by sympodial positions 1, 2 and >2 within zones<br/>as divided by nodes during open boll (100 days after emergence) at<br/>Tennesse Valley Research Extension Center.

<sup>z</sup> Means within columns followed by the same letter are not different according to the Least Significant Difference means separation test  $P\alpha$ =0.05.

<sup>y</sup> Pos=Sympodial (Fruiting) Position.

<sup>x</sup> Zone 1 retention nodes 5 to 9; Zone 2 retention nodes 10 to 14; Zone 3 retention nodes 15 to 19.

<sup>w</sup> Average six plants with normal terminal sampled destructively per plot.

Table 3.20 Measure of cotton maturity (nodes above white flower) [NAWF], (nodes above cracked boll) [NACB], (node of last harvestable boll)<sup>x</sup> [NLHB] and percent open boll as affected by nematicides during bloom (70 DAE<sup>w</sup>) and open boll (100 DAE<sup>w</sup>) in *R. reniformis* infested soils at Mississippi State University.

Treatment	NAWF	NACB	NLHB	Open Boll
	(bloom)	(open boll)	(open boll)	(%) <sup>y</sup>
Untreated	8.1ab <sup>z</sup>	8.2a	16.0a	22.0d
Temik 15G	7.6bc	8.4a	17.0a	24.0c
Aeris®	8.1ab	8.3a	16.0a	24.0c
Aeris <sup>®</sup> + Votivo <sup>®</sup>	8.3a	8.5a	16.0a	21.0d
Temik <sup>®</sup> + Vydate	7.4cd	7.4bc	16.0a	29.0a
C-LV®				
Aeris <sup>®</sup> + Vydate	7.0d	7.7b	16.0a	26.0b
C-LV®				
Aeris <sup>®</sup> + Votivo <sup>®</sup>	7.1cd	7.1c	16.0a	30.0a
+ Vydate C-LV®				
LSD (0.05)	0.42	0.37	0.76	1.4

<sup>z</sup> Means within columns followed by the same letter are not different according to the Least Significant Difference means separation test  $P\alpha$ =0.05.

<sup>y</sup> % open boll derived from number of open first position bolls/total number of first position bolls retained.

<sup>x</sup> Average six plants with normal terminal sampled destructively per plot.

<sup>w</sup> DAE=Days After Emergence.

Table 3.21Measure of cotton maturity (nodes above white flower) [NAWF], (nodes<br/>above cracked boll) [NACB], (node of last harvestable boll)<sup>x</sup> [NLHB] and<br/>percent open boll as affected by nematicides during bloom (70 DAE<sup>w</sup>) and<br/>open boll (100 DAE<sup>w</sup>) in *R. reniformis* infested soils at Tennessee Valley<br/>Research Extension Center.

Treatment	NAWF	NACB	NLHB	% Open Boll <sup>y</sup>
Untreated	10.1ab <sup>z</sup>	10.4a	19.0a	10.0d
Temik 15G	10.1ab	9.3ab	18.3b	24.0b
Aeris®	10.1ab	10.4a	18.3b	10.0d
Aeris <sup>®</sup> + Votivo <sup>®</sup>	10.3a	10.0a	17.2c	16.0c
Temik 15G +	9.7abc	8.5b	17.3c	30.0a
Vydate C-LV <sup>®</sup>				
Aeris <sup>®</sup> + Vydate	10.1ab	9.7ab	17.7c	22.0b
C-LV®				
Aeris <sup>®</sup> + Votivo <sup>®</sup>	9.5c	9.7ab	17.5c	25.0b
+ Vydate C-LV <sup>®</sup>				
LSD (0.05)	0.6	0.6	0.5	1.3

<sup>z</sup> Means within columns followed by the same letter are not different according to the Least Significant Difference means separation test  $P\alpha$ =0.05.

<sup>y</sup> % open boll derived from number of number of open first position bolls/total number of first position bolls retained.

<sup>x</sup> Average six plants with normal terminal sampled destructively per plot.

<sup>w</sup> DAE=Days After Emergence.

Table 3.22Monopodial (vegetative) branch and boll production<sup>x</sup> at Mississippi State<br/>University collected during open boll (100 days after emergence) to<br/>showcase overall plant performance treated with nematicides *R. reniformis*<br/>infested soils.

Treatment	Monopodial Branch/Plant <sup>y</sup>	Monopodial Bolls/Plant
Untreated	1.8c <sup>z</sup>	2.0c
Temik 15G	2.1bc	3.0ab
Aeris®	1.8c	2.0bc
Aeris <sup>®</sup> + Votivo <sup>®</sup>	2.0d	2.0bc
Temik 15G + Vydate C-LV <sup>®</sup>	3.0ab	3.0a
Aeris <sup>®</sup> + Vydate C-LV <sup>®</sup>	3.0a	3.0ab
Aeris <sup>®</sup> + Votivo <sup>®</sup> + Vydate	2.0ab	3.0ab
C-LV®		
LSD (0.05)	0.4	0.7

<sup>z</sup> Means within columns followed by the same letter are not different according to the Least Significant Difference means separation test  $P\alpha$ =0.05.

<sup>y</sup> Monopodial branch and boll production represents plant health.

<sup>x</sup> Average six consecutive plants with a normal terminal sampled destructively.

Table 3.23Monopodial (vegetative) branch and boll production<sup>x</sup> at Tennessee Valley<br/>Research Extension Center collected during open boll (100 days after<br/>emergence) to showcase overall plant performance treated with nematicides<br/>in *R. reniformis* infested soils.

Treatment	Monopodial Branch/Plant <sup>y</sup>	Monopodial Bolls/Plant
Untreated	0.4ab <sup>z</sup>	1.0a
Temik 15G	1.4a	2.3a
Aeris®	0.9a	2.0a
Aeris <sup>®</sup> + Votivo <sup>®</sup>	0.3b	2.2a
Temik 15G + Vydate C-LV <sup>®</sup>	1.3a	2.4a
Aeris <sup>®</sup> + Vydate C-LV <sup>®</sup>	1.0a	2.8a
Aeris <sup>®</sup> + Votivo <sup>®</sup> + Vydate	1.4a	2.3a
C-LV®		
LSD (0.05)	0.8	0.7

<sup>z</sup> Means within columns followed by the same letter are not different according to the Least Significant Difference means separation test  $P\alpha$ =0.05.

<sup>y</sup> Monopodial branch and boll production represents plant health.

<sup>x</sup> Average six consecutive plants sampled destructively per plot.

Table 3.24Basal stalk and boll diametersw (70 days after emergence) taken at ninth and<br/>twelth node below terminal to showcase improved plant performance<br/>resulting from nematicide treatments in *R. reniformis* infested soils at<br/>Mississippi State University.

Treatment	Basal Stalk Diameter	Boll D (m	Boll Diameter Difference	
	(mm) <sup>y</sup>	Node-9 <sup>x</sup>	Node-12 <sup>x</sup>	(mm)
Untreated	5.9d <sup>z</sup>	24.8d	32.9bc	8.2d
Temik 15G	9.0ab	29.5b	32.9bc	3.5a
Aeris®	6.9c	27.8c	32.4bc	7.0c
Aeris <sup>®</sup> + Votivo <sup>®</sup>	8.4b	28.4c	32.8bc	5.9b
Temik 15G + Vydate C-	9.8a	31.0a	33.6b	2.6a
LV®				
Aeris <sup>®</sup> + Vydate C-LV <sup>®</sup>	7.0c	25.5d	32.9bc	5.1b
$Aeris^{\mathbb{R}} + Votivo^{\mathbb{R}} +$	9.7a	30.5a	34.2a	2.3a
Vydate C-LV <sup>®</sup>				
LSD (0.05)	0.9	1.1	0.6	1.1

<sup>z</sup> Means within columns followed by the same letter are not different according to the Least Significant Difference means separation test  $P\alpha$ =0.05.

<sup>y</sup> Diameters taken with digital calipers at boll center and cotyledonary node for stalk.

<sup>x</sup> Bolls at node 12 from terminal are the oldest boll and bolls at node 9 are the youngest.

<sup>w</sup> Six consecutive plants sampled destructively per plot.

Table 3.25Basal stalk and boll diametersw (70 days after emergence) at ninth and<br/>twelth node below terminal to showcase improved plant performance<br/>resulting from nematicide treatments in *R. reniformis* infested at Tennessee<br/>Valley Research Extension Center.

Treatment	Basal Stalk Diameter	Boll D (n	Boll Diameter Difference	
	(mm) <sup>y</sup>	Node-9 <sup>x</sup>	Node-12 <sup>x</sup>	(mm)
Untreated	6.3b <sup>z</sup>	9.2d	26.4d	17.2c
Temik 15G	8.6ab	15.3b	31.3b	15.9b
Aeris®	7.5ab	15.5b	31.0b	15.5b
Aeris <sup>®</sup> + Votivo <sup>®</sup>	7.2ab	15.4b	31.1b	15.8b
Temik 15G + Vydate C- LV <sup>®</sup>	9.4a	19.1a	32.1a	13.0a
Aeris <sup>®</sup> + Vydate C-LV <sup>®</sup>	8.0ab	13.4c	26.1d	13.9a
$Aeris^{\mathbb{R}} + Votivo^{\mathbb{R}} +$	8.0ab	15.3b	29.2c	12.7a
Vydate C-LV <sup>®</sup>				
LSD (0.050)	1.9	0.4	1.7	1.8

<sup>z</sup> Means within columns followed by the same letter are not different according to the Least Significant Difference means separation test  $P\alpha$ =0.05.

<sup>y</sup> Diameters taken with digital calipers at boll center and cotyledonary node for stalk.

<sup>x</sup> Bolls at node 12 from terminal are oldest boll and bolls at node 9 are youngest.

<sup>w</sup> Average six consecutive plants sampled destructively per plot.

Table 3.26Yield of Phy 375 in pounds (Lbs.) lint cotton per acre treated with<br/>nematicides grown in *R. reniformis* infested soils at MSU and TVREC.

	Lbs Lint/Ac <sup>y</sup>	Lbs Lint/Ac
Treatment	MSU	TVREC
Untreated	1418.0e <sup>z</sup>	582.0f
Temik 15G	1529.0bcd	1168.0c
Aeris®	1474.0d	783.0e
Aeris <sup>®</sup> + Votivo <sup>®</sup>	1483.0d	887.0d
Temik 15G + Vydate C-LV <sup>®</sup>	1755.0a	1331.0a
Aeris <sup>®</sup> + Vydate C-LV <sup>®</sup>	1557.0b	1246.0b
Aeris <sup>®</sup> + Votivo <sup>®</sup> + Vydate	1610.0a	1328.0a
C-LV®		
LSD (0.05)	65.1	46.1

<sup>z</sup> Means within columns followed by the same letter are not different according to the Least Significant Difference means separation test  $P\alpha$ =0.05.

<sup>y</sup> Lbs lint cotton formulated using harvested seed cotton weights x established lint % for Phy 375 taken from MSU Official Variety Trials (OVT).

Treatment	Nematode	Juvenile	Egg	Shoot Biomass	Root
	Population <sup>u</sup>	number/500	number/500	(Grams) <sup>y</sup>	Biomass
		cc <sup>v,w</sup>	cc <sup>v,w</sup>		(Grams) <sup>y</sup>
Untreated	0.0	0.0g <sup>z</sup>	0.0d	48.0fgh	46.6d
Temik 15G	0.0	0.0g	0.0d	68.5ab	52.5b
Aeris®	0.0	0.0g	0.0d	70.3a	55.6a
Aeris <sup>®</sup> +	0.0	0.0g	0.0d	64.5bc	57.0a
Votivo®					
Untreated	2,500	1,597.0fg	1,123.0cd	46.9gh	35.0f
Temik 15G	2,500	901.0fg	438.0d	70.2a	51.6b
Aeris®	2,500	7,892.0c	4,282.0ab	56.0d	46.7d
Aeris <sup>®</sup> +	2,500	1,597.0fg	5,214.0a	60.9c	49.7c
Votivo®		_			
Untreated	5,000	3,901.0e	1,975.0cd	45.7h	34.5f
Temik 15G	5,000	1,087.0f	1,306.0cd	62.6c	51.4b
Aeris®	5,000	5,021.0de	2,639.0bc	51.7ef	45.3d
Aeris <sup>®</sup> +	5,000	9,754.0b	5,163.0a	53.3de	45.6d
Votivo®					
Untreated	7,500	5,995.0d	1,442.0cd	41.4i	25.1g
Temik 15G	7,500	1,576.0f	1,391.0cd	52.9de	51.4b
Aeris®	7,500	4,172.0e	1,759.0cd	46.2h	39.8e
Aeris <sup>®</sup> +	7,500	5,459.0d	2,820.0bc	50.6efg	44.6d
Votivo®				-	
LSD (0.05)		1236.0	2196.5	4.1	2.7
	• 1 01			1:00	

Table 3.27Effect of nematicides on reproduction of *R. reniformis* and shoot and root<br/>biomass development of Phy 375 under varying *R. reniformis* populations<br/>grown under greenhouse environments at 90 days after emergence.

<sup>z</sup> Means within columns followed by the same letter are not different according to the Least Significant Difference means separation test  $P\alpha$ =0.05.

<sup>y</sup> Shoot and root biomass were acquired from the one plant grown in a 3.0 inch pot.

<sup>x</sup> Two seed per pot planted 0.5 inches deep and one removed after emergence.

<sup>w</sup> 3.0 inch pot represented 500 cc of soil.

<sup>v</sup> Juvenile and eggs of *R. reniformis* extracted from the 500 cc of soil via elutriator and centrifuge process.

<sup>u</sup> *R. reniformis* added to soil at planting using a pipette via a graduated factor.

		NEED		D1 . TT 1 1	10.00
Treatment	Nematode	NFFB	Total Node	Plant Height	HNR
	Population <sup>v</sup>	(number) <sup>y</sup>	(number)	(inch)	(inch)
Untreated	0 <sup>z</sup>	6.0d <sup>z</sup>	12.0c	21.4de	1.7bc
Temik 15G	0	7.0c	13.0b	23.0abc	1.7bc
Aeris®	0	6.0d	13.0b	23.4ab	1.8ab
Aeris <sup>®</sup> +	0	6.0d	14.0a	23.8a	1.7bcd
Votivo®					
Untreated	2,500	7.3b	11.0d	16.0g	1.4e
Temik 15G	2,500	6.0d	13.0b	23.4ab	1.8ab
Aeris®	2,500	7.0b	12.0c	22.3bcd	1.8ab
Aeris <sup>®</sup> +	2,500	7.0b	14.0a	23.2abc	1.7cd
Votivo®					
Untreated	5,000	8.0a	10.0d	14.0h	1.3f
Temik 15G	5,000	7.0b	12.0c	22.6abc	1.9a
Aeris®	5,000	7.0b	12.0c	21.0e	1.7cd
Aeris <sup>®</sup> +	5,000	7.0b	13.0b	22.0cd	1.7bcd
Votivo®					
Untreated	7,500	8.0a	10.0e	14.4gh	1.4e
Temik 15G	7,500	7.0b	12.0c	21.3de	1.8bc
Aeris®	7,500	7.0b	13.0b	19.2f	1.6d
Aeris <sup>®</sup> +	7,500	7.0b	12.0c	19.0f	1.4e
Votivo®					
LSD (0.05)		0.1	0.4	1.2	0.1

Table 3.28Effect of nematicides on growth of Phy 375 WRF grown under varying *R. reniformis* populations under greenhouse environments<sup>w,x</sup> at 90 days after emergence.

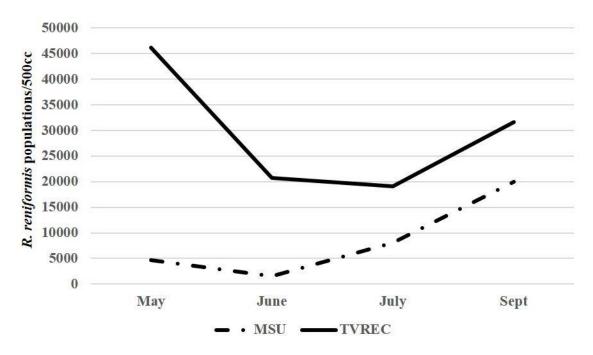
<sup>z</sup> Means within columns followed by the same letter are not different according to the Least Significant Difference means separation test  $P\alpha$ =0.05.

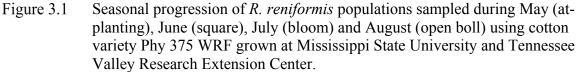
<sup>y</sup> Growth parameters were acquired from the one plant grown in a 3.0 inch pot.

 $\times$  3.0 inch pot represented 500 cc of soil.

<sup>w</sup> Two seed per pot planted 0.5 inches deep and one removed after emergence.

<sup>v</sup> *R. reniformis* added to soil at planting using a pipette via a graduated factor.





- <sup>z</sup> Samples acquired on a per plot basis and averaged across all plots on per 500 cc basis to display population dynamics of *R. reniformis* at each cotton growth stage.
- <sup>y</sup> Six samples per plot were acquired using a fluted probe from six inches from the row middle in a manner to obtain three samples from each of the two row plots.
- <sup>x</sup> Sample depth was approximately three inches deep.
- <sup>w</sup> Samples were bagged and cooled away from direct sunlight until sampled using the elutriator/centrifuge system.

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#### CHAPTER IV

### PERFORMANCE OF COMMERCIALLY AVAILABLE *GOSSYPIUM HIRSUTUM* VARIETIES GROWN IN *ROTYLENCHULUS RENIFORMIS* INFESTED SOILS WITH AND WITHOUT NEMATICIDES

#### Abstract

Reniform nematode (Rotylenchulus reniformis Linford and Oliveira) infests 36% of the Mississippi cotton (Gossypium hirsutum) acres promoting national economic losses of 58,000 bales of G. hirsutum in 2015. Previously nematodes were managed using an at-planting treatment of Temik 15G or soil fumigants. With label loss of Temik 15G and expense of soil fumigants need arose to develop an integrated nematode management program entailing understanding which commercial G. hirsutum varieties exhibit tolerance to *R. reniformis*. Tolerance to root knot nematode (*Meloidogyne ingognita*) exists, but little tolerance to R. reniformis has been observed in G. hirsutum varieties. However, research indicates some varieties grow and yield better than other varieties in R. reniformis infested soils. Studies at Mississippi State University (Mississippi State, MS) during 2012 indicated all varieties evaluated had improved growth, development and yield with the addition of a nematicide. Greenhouse and field studies indicated some commercially available varieties grew and yielded better than others when grown without nematicides. Evaluated commercial varieties, Stv 5458 (Bayer Crop Science-Raleigh, NC), FM 1740 B2RF (Bayer Crop Science, Lubbock, TX) and Phy 499 WRF (Dow Agro 102

Science, Indianapolis, IN) were comparable in yield when untreated compared to those treated with a nematicide. Phy 375 (Dow Agro Science-Indianapolis, IN) responded positively to a nematicide treatment. Response differences based on soil type indicated positioning a variety by soil type preference can improve performance in *R. reniformis* infested soils. Greenhouse studies at Mississippi State University indicated all varieties had improved root and shoot growth using a nematicide. As *R. reniformis* populations increased, a reduction in shoot and root growth was observed, but performance varied by variety.

#### Introduction

Reniform nematode (*Rotylenchulus reniformis* Linford and Oliveira) has become the most damaging pathogen to cotton (*Gossypium hirsutum*). *R. reniformis*, first described in 1931 (Linford and Oliveira, 1940), is a tropical and subtropical pest present throughout the United States *G. hirsutum* producing regions (Heald and Robinson, 1990; Kinloch and Sprenkel, 1994; Star, 2007; Koenning et al., 1999). Since 1960, *R. reniformis* has shown an adaptive capability to survive colder environments allowing colonization of much of the eastern half of the *G. hirsutum* belt (Heald and Robinson, 1990) and as far north as Lubbock, Texas and the Missouri bootheel (Heald and Thames, 1982; Wrather et al., 1992). Today, *R. reniformis* has been identified and associated with 7% annual *G. hirsutum* yield loss totaling \$130 million in Mississippi, Alabama, Tennessee, Texas, Missouri, Florida, North Carolina, Louisiana, South Carolina, Arkansas and Georgia (Blasingame et al., 2009; Koenning et al., 1999). In Mississippi, an annual yield loss of 235,398, 252,023 and 56,378 bales occurred in 2004, 2005 and 2011, respectively (Blasingame, 2004; 2005; 2011). By 2002, more than 32% of *G*. *hirsutum* acreage in Mississippi were infested with *R. reniformis* causing a 5.5% yield reduction (Lawrence et al., 2002). Gazaway and McLean (2003) reported *R. reniformis* infested more than 36% of Alabama *G. hirsutum* acreage and was increasing.

Since 2004, the *G. hirsutum* industry began moving away from the granular, atplanting treatment with Temik 15G for nematode management. Previously, Temik 15G was the main-stay for nematode management in *G. hirsutum*. Since use of Temik 15G on *G. hirsutum* was removed from the product label, producers had to find alternate control methods involving a complete integrated nematode management program with limited reliance on chemicals for *R. reniformis* management. A portion of this integrated approach involves identifying strengths and characteristics of currently available varieties grown in *R. reniformis* infested soils.

Presently, there are no *G. hirsutum* varieties marketed as *R. reniformis* resistant, but much effort is being directed toward resolving this need (Usery et al., 2005; Robinson et al., 2007; Starr, et al., 2007). Some varieties have been shown to possess nematode tolerance (Usery et al., 2004; 2005) at low to moderate nematode populations (Starr et al., 2007; Weaver et al., 2007). Gene identification driving nematode resistance in *G. hirsutum* has made positive strides. Davis (2011) reported root knot nematode (*Meloidogyne incognita*) resistance is a multi-gene trait difficult to maintain in breeding programs. Bell and Robinson (2004) reported resistance to *R. reniformis* requires introgression of genes from *G. longicalyx*. They further reported DNA markers imparting resistance to *M. incognita* and *R. reniformis* had been identified. The United States Department of Agriculture released two varieties (LONREN-1 and LONREN-2) originating from *G. longicalyx* (a wild *Gossypium* species from Africa) that are very resistant to R. reniformis (Usery et al., 2005; Robinson et al., 2007; Starr, et al., 2007). Percival et al. (1999) and Yik and Birchfield (1984) cited G. longicalyx as having complete resistance to *R. reniformis* preventing females entering the root from forming their normal reniform shape. This reduces normal sexual activities which prevents egg production and subsequent generations. However, G. longicalvx cultivars have serious commercial limitations. They have a poor growth habit despite being well adapted to dry and high saline environments and having excellent lint properties. A problem of LONREN cultivars is susceptibility to root borne fungi, and they can support only low populations of *R. reniformis* in the greenhouse or field (Bell et al., 2009; Bell et al., 2011; Weaver et al., 2011; Weaver et al., 2013). Where R. reniformis populations ranged from 10,000 to 50,000 per 100 cm<sup>3</sup> of soil at planting, LONREN lines were intolerant having smaller root systems, stunting and reduced yields (Nichols et al., 2010; Sikkens et al., 2011). Rotylenchulus reniformis control in LONREN lines is by hypersensitive reactions where root tissues damaged upon infection promotes *R. reniformis* death, but negative plant effects between radical emergence and full seedling growth does occur (Sikkens et al., 2011; Weaver et al., 2013). Schrimsher, et al. (2014) showed nematicides could aid in *R. reniformis* management during this susceptible period. A further issue with G. *longicalyx* cultivars is incompatibility with G. *hirsutum* due to chromosome differences; G. hirsutum (2n=52, similar to other Gossypium sp.) and G. longicalyx (2n=26) (Bell, 1984). Genetic markers have been identified, leading to successful breeding programs involving *R. reniformis* resistance (Avila et al., 2005; 2006; Avila and Stewart, 2008; Dighe et al., 2005; Bell and Robinson, 2004; Robinson and Bell, 2006; Robinson et. al. 2007; 2004; 1997; Bell et al., 2009; Jenkins et al., 2004; Young et al., 2004). Robinson et

al. (2007) has been successful in introgressing resistance to *R. reniformis* into upland cotton from *G. longicalyx*.

BARBEN is another cotton cultivar derived from an exotic cotton species, *G. barbadense*. Through years of searching *G. barbadense* cultivars, tolerance to *R. reniformis* was discovered which reduced egg production to as low as 8% and thus, subsequent generations (Yik and Birchfield, 1984; Robinson and Percival, 1997; Robinson et al., 2004). Robinson et al. (2004) found an accession, GB-713, which reduced egg production of *R. reniformis* to as low as 3% and is now being used to introgress resistant genes into *G. hirsutum*. In 2012, the USDA, Mississippi State University and Cotton Incorporated launched BARBEN-713. Sikkens et al. (2012) reported this cultivar suppressed reproduction resulting in low levels of *R. reniformis* with yields comparable to commercial cotton cultivars. These results show promise relative to possibilities of crossing BARBEN-713 with high yielding, commercially available varieties. A suitable commercial variety possessing resistance is still years away and a need exists to better understand how commercial varieties perform in a *R. reniformis* infested environment.

Numerous studies have been conducted evaluating performance of commercial varieties in nematode infested soils. Since 1988, eleven breeding lines tolerant to *M. incognita* have been released (Jones et al., 1988; Cook et al., 1997 a and b; Cook and Robinson, 2005). These varieties yield well in *M. incognita* infested fields of their production regions. However, according to Koening et al. (2001), these varieties may have geographic limitations. Wheeler, et al. (2014) reported a positive economic interaction between nematicides plus foliar application of Vydate C-LV<sup>®</sup> and variety in

*M. incognita* infested fields. Usery et al. (2004; 2005) and Legee et al. (2007) reported several varieties had tolerance to high R. reniformis infestation using plant mapping to evaluate variety performance in these environments. Luangkhot et al. (2015) further reported currently grown non-tolerant varieties responded to nematicides in greenhouse and field environments where *R. reniformis* was present. This group did not use plant mapping to evaluate variety strengths to understand variety management in this environment. Usery et al. (2004; 2005) reported that early maturing varieties had greater R. reniformis tolerance resulting in higher yields and lower nematode feeding. Further work evaluating commercial variety performance in nematode infested soils was reported by Phipps and Eisenback (2005) and Davis (2005) as related to *M. incognita* and Sciumbato et al. (2005) as related to R. reniformis, indicating no difference among Gossypium hirsutum maturity groups. Koenning et al. (2005), however, reported late maturing varieties performed better than early maturing varieties in soils infested with Columbia lance nematode (Hoploaimus columbus). Williams et al. (2004) reported similar findings with *M. incognita*. Phipps and Eisenback (2005) further reported net dollar return was greater when using tolerant G. hirsutum varieties planted in M. *incognita* infested fields. They also reported nematicides were still economically beneficial when used with tolerant varieties. There are several commercial varieties that show promise in highly infested nematode soils (Davis et al., 2010). McPherson and Rush (2011) reported PHY 367 had excellent growth and yield in *M. incognita* infested soils despite not being treated with Telone or Temik 15G.

Today, Fiber Max and Stoneville, subsidiaries of Bayer Crop Science are discussing potential tolerance to nematodes, especially *R. reniformis*. There is a need to

understand the fruiting mechanisms and performance of new, high yielding *G. hirsutum* varieties in nematode infested soils. With cost of *G. hirsutum* seed and technology, it is important to minimize controllable risks. In addition, new cotton varieties containing advanced technologies retain higher fruit levels and produce higher yields than older technologies and varieties which makes minimizing events that limit yield important (Stewart and Smith, 2007). Also, with label removal of Temik 15G, identifying *R. reniformis* tolerant varieties can greatly improve *G. hirsutum* performance and improve efficacy of seed treatment nematicides. The best method to understand how a variety fits a *R. reniformis* management system is by establishing and understanding fruiting architecture using plant mapping (Jenkins and McCarty, 1995; Kerby et al., 1987; Smith et al., 1996; 1998). Plant mapping determines growth propensity and fruit retention with environmental and pest interactions. In addition, greenhouse evaluations must occur concurrently to establish an innoculated population (*Pi*) tolerance for each variety tested.

The study objective was to evaluate and map growth, development and yield of five *Gossypium hirsutum* varieties grown with and without nematicide treatments in *R*. *reniformis* infested soils to determine tolerance among commercial *G. hirsutum* varieties.

#### **Materials and Methods**

#### In-field variety treatments (with and without nematicides)

Two studies were conducted at R. R. Foil Plant Science Research Center at North Farm of Mississippi State University Mississippi State, MS on two different soil types containing an established population of *R. reniformis*. Soil tests were conducted prior to planting and analyzed at Mississippi State University Extension Soil Testing Lab. At location one, the soil was a Marrietta fine sandy loam and at location two, the soil type was a Leeper silty clay loam.

Five commercially available varieties (FM 1740 B2RF, Stv 5458 B2RF, Stv 5288 B2RF, Phy 375 WRF and Phy 499 WRF) with and without NST, Aeris<sup>®</sup> (Bayer Crop Science, Raleigh, NC) (Table 4.1) were evaluated. Planting occurred on May 15 and 16, 2012, using a four-row Almaco cone planter (Allan Manufacturing Company, Nevada, IA) using seed previously treated with Aeris<sup>®</sup> by Bayer Crop Science and counted prior to planting through a Seed Counter Model U (International Market and Design Corporation, San Antonio, TX) to deliver consistent seed per plot. Weed control consisted of applications of Power Max<sup>®</sup> (glyphosate) (Monsanto, St. Louis, MO) overthe-top of cotton at 1.0 lb ai/Ac followed by a lay-by application of Karmex DF <sup>TM</sup> (diuron) (DuPont USA, Wilmington, DE) at 1.0 lb ai/Ac. Both trial locations had furrow irrigation available, but was not used due to adequate rainfall.

#### **Experimental design and establishment**

Trial design used at both locations was a randomized complete block (RCB) design with five replications. This statistical method was selected to address the spatial distribution of nematodes across the field thereby reducing variability of nematode populations existing between plots. Data was analyzed using Analysis of Variance (ANOVA) for a 5 by 2 Factorial with a RCB factor (ARM 8 Statistical Software, Gylling Data Management, Brookings, SD) where block and treatment effects were evaluated to minimize dgree of error and improve confidence intervals among experimental units. Means were separated using the Least Significant Difference (LSD) test at  $P\alpha$ =0.05. Individual plot length consisted of two-row plots of 50 feet with 10 foot alleys. Row 109 spacing consisted of a solid planting pattern planted on 38 inches centers with a seeding rate of 4.0 seed per row feet. Border effects were reduced by planting border rows with additional cotton and using a solid planting pattern. The lack of bordering in the front and back of the trial area was compensated for by acquiring samples from within the plots to avoid edge effects.

#### In-season evaluation prior to fruiting

In-season field evaluations at both locations included vigor and plant population followed by an extensive plant mapping program where six consecutive plants consisting of a normal terminal were cut at the ground level, tagged and removed to be monitored via plant mapping processes for boll retention and growth (Gutherie and Kerby, 1993).

#### Evaluation of vigor, plant population and hypocotyl lengths

Visual plant vigor and plant population were evaluated at 14 days following emergence (DAE). Vigor was established using two processes; visual assessment on a scale of one to five where one had best vigor and five lowest vigor, based on overall plant growth and health and hypocotyl measurement. Hypocotyl measurement involved a measurement of length from seed embryo axis to cotyledonary node. The hypocotyl length is a direct measurement of seedling vigor and energy stored in the seed. Furthermore, hypocotyl measurements, as opposed to visual evaluations, provide a quantifiable and accurate method to analyze vigor (Legee and Smith, 2002). Plant population was determined by counting every plant in all plots to determine plants per hectare.

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#### **In-season evaluation during fruiting**

Evaluation during mid-square growth stage

In-season evaluation occurred at mid-square, bloom and open boll growth stages. The first plant mapping occurred at mid-square. Evaluation criteria included: plant height (PH) (from the cotyledonary node to terminal), node of first fruiting branch (NFFB), total nodes (TN) (cotyledonary node treated as zero), height to node ratio (HNR), fruit retention by position along sympodial (fruiting branch) and average plant height by node measurements. Average plant height by node measurements were conducted by measuring each internode length separately from cotyledons to terminal leaf 0.5 inch in size and internode lengths summed to obtain final plant height (Kerby, et al., 2003). This method facilitated collecting final height and individual internode growth across time as affected by *G. hirsutum* variety in presence of *R. reniformis*. Six consecutive plants possessing a normal terminal were destructively sampled per plot totaling 30 plants sampled. Evaluation time was two weeks following square initiation.

#### Evaluation during bloom growth phase

Evaluation criteria on six consecutive plants per plot included the following: PH in inches, TN, nodes above white flower (NAWF), node of white flower (NOWF), fruit retention by position as stated previously and HNR in mm. In addition, caliper (General Ultra Tech, Port Washington, NY) was measured at the cotyledonary node to obtain basal stalk diameter and from unopened first position bolls at nodes 9 and 12 below the terminal to determine treatment effect on boll size. Bolls at the ninth node below the terminal represented younger bolls while bolls at the twelfth node represented older bolls. Evaluation occurred during late bloom.

#### Evaluation during open boll growth phase

Evaluation during open boll plant growth stage included the following criteria on six consecutive plants: PH, TN, HNR, nodes above cracked boll (NACB), fruit retention by position and percent (%) open boll. Monitoring began when cotton bolls of the earliest treatment in the study were approximately 30% open collectively, based on visual assessment.

#### Machine harvest

Defoliation was conducted based on visual assessments of 60% open boll with harvest aids applied using high clearance ground equipment. Harvest was conducted using a small plot machine harvester (John Deere 9965, Moline, IL) equipped with a weighing system (Rice Lake 9201i, Rice Lake Weighing Systems, Rice Lake, WI) to measure seed cotton of individual plots during harvest. Seed cotton weights were converted to lint pounds per acre using historical lint percentages established via University Official Variety Trials at Mississippi State University (Mississippi State, MS).

#### *R. reniformis* sampling and processing

*Rotylenchulus reniformis* collection included nematode soil samples collected prior to planting from each plot to establish an initial population density. Nematode populations were monitored at-planting, square, bloom and open boll. Core soil samples were acquired using a fluted probe designed to collect multiple samples per plot. Probe dimensions were 3.44 inches at the top and tapering to 0.75 inches at the bottom facilitating multiple samples without loss of soil. Length of sample device was 11.0 inches to guarantee acquisition of 500 cc of soil. Samples were acquired from the side of the emerged row at a distance of about six inches in a zig-zag pattern allowing six samples to be acquired at three samples per row. Sampling was conducted at an approximate depth of four inches. The sampling process was always conducted when the soil possessed adequate moisture levels and preferably at field capacity since the R. *reniformis* move deeper into the soil profile as soil dries. Samples were bagged in plastic bags and kept in cold storage (35°F) until extraction using a semi-automatic elutriator (W.S. Tyler Co, Mentor, OH) and centrifugal flotation (1 EC Model K Centrifuge, Needham Hts, MA) (Byrd et al., 1976). Rotylenchulus reniformis extraction process was as follows: Collected soil on an individual plot basis was placed into a 450 ml beaker and processed through a 60 mesh screen followed by a 400 mesh screen using the aqueous extraction process of the elutriator. Soil was removed and placed into a 250 ml beaker, water drained and the sample poured into centrifuge tubes where it contained 1.0 inch of soil and was spun for six minutes at about 2,500 RPM. Excess water was removed and mixed with a sucrose mixture (454 g sucrose per 1,000 ml of water) followed by a one minute process in the centrifuge. The liquid was poured through a 500 mesh screen and sample refrigerated (35°F) until counted. The resulting nematodes were enumerated using a stereo-microscope (Nikon AFX-11A, Minato-ku, Tokyo).

# *R. reniformis* tolerance of commercially available cotton varieties grown at varying populations under greenhouse environments

Study establishment and experimental design

To support field findings, two seperate greenhouse studies were established using five cotton varieties Phy 375, Phy 499, Stv 5288, Stv 5458 and FM 1740 treated with Aeris<sup>®</sup> or untreated (Table 4.2). Varieties were planted at two seeds per 3.0 inch clay pot

into a sterile soil. Soil medium included an autoclave, fine sandy loam. All pots were brought to the same level to ensure 500 cc. Planting depth for all seed was 0.5 inch. Upon emergence, one plant was removed to leave one plant per container. Nematode populations were applied in a liquid solution to the soil using a graduated pipette and included 0, 2,500, 5,000 or 10,000 *R. reniformis* per 500 cc of soil. Each study was conducted for 90 days. Experimental design was a RCB design using four replications. Data were analyzed via ANOVA for a RCB (ARM 8 statistical software) where block and treatment effects were evaluated to minimize dgree of error and improve confidence intervals among experimental units. Means were separated using the LSD at  $P\alpha$ =0.05 level of probability.

#### Evaluation criteria

Before harvest evaluations included TN, PH, NFFB, HNR and basal stalk diameter. At harvest evaluations included root and shoot biomass and nematode extraction (eggs and juveniles). At harvest, shoot biomass was separated from the root biomass using hand pruners. The shoot was weighed and mass recorded. Roots were extracted from the soil in a bucket. Soil-free roots were soaked and stirred in 10% bleach solution for three minutes and roots weighed. Remaining solution was poured through 250 over 500 mesh screen to obtain egg numbers. The remaining soil was mixed with 1,000 ml of water and processed through a 60 over 325 mesh screen and centrifuged for six minutes at 2,500 rpm. Excess water was removed and mixed with a sucrose mixture (454 g sucrose per 1,000 ml of water) followed by a one minute centrifuge at 2,500 rpm. The liguid was poured through a 500 mesh screen and sample refrigerated in a 250 ml beaker until counted. Nematode numbers were counted under a stereo microscope for *R*. *reniformis* juveniles and eggs by pipetting 20 ml of liquid into a quadrated petri dish.

#### **Results and Discussion**

#### Seasonal population development of the reniform nematode

*Rotylenchulus reniformis* population progression across time becomes important in determining impact on growth and development of *G. hirsutum* at each growth stage. Relating nematode numbers to root development has aided in establishing effective treatments against *R. reniformis* nematode populations resulting in greater root development at season end (Lawrence and McLean, 1995 a and b; 1996 a and b).

At the Marrietta fine sandy loam location, *R. reniformis* population was low during May (at-planting) evaluation and remained unchanged until June (square) when the population began trending upward (Table 4.3 and Fig. 4.1). The largest population increase occurred between July (bloom) and August (open boll) at this location (Table 4.3). At the Leeper silty clay loam location, initial population development followed a similar pattern between May and June, but from June to August began a rapid population increase (Table 4.3 and Fig. 4.1). Major population increases began one month earlier at location one relating to pre-square to late-bloom growth phases.

# Variety influence grown in *R. reniformis* infested soils with and without nematicides prior to fruiting

#### **Plant Population**

Plant populations were similar for treated or untreated seed within each variety with the exception of FM 1740 where treating the seed resulted in a reduced population (Table 4.4). However, Stv 5458 populations were greater compared to varieties,

regardless of seed treatment. In this location, varieties possessing the lowest number of emerged plants included Aeris<sup>®</sup> treated FM 1740 and Phy 375 when compared to Stv 5458. Plant population at the Leeper silty clay loam location was reduced in all treated seeds except FM 1740 and Stv 5458 when compared within varieties (Table 4.5). Untreated Phy 499 followed by Stv 5458 (Aeris<sup>®</sup> treated and untreated) and untreated Phy 499 were greater in plant population than remaining treatments. Early maturing cotton varieties, Phy 375 and Stv 5288, were improved in plant population without addition of a nematicide. FM 1740 was lower in overall population compared to Phy 499 and Stv 5458 and was not improved with the addition of nematicide (Table 4.5).

#### Vigor evaluation

Seedling vigor at the Marrietta fine sandy loam location (Table 4.4) was improved in Phy 499 when using a nematicide while nematicide had no affect on seedling vigor in the other varieties. The variety with greatest vigor was untreated Phy 499 at the Marrietta fine sandy loam location and vigor was reduced using a nematicide (Phy 499 is a late maturing variety which does not exhibit rapid early growth). Seedling vigor of varieties grown at the Leeper silty clay loam location (Table 4.5) was again reduced with addition of a nematicide except with Stv 5458 where seed treatment had no influence. Differences in variety seedling vigor between treated and untreated were not as great at the Leeper silty clay loam location indicating a possible interaction of location and *R. reniformis*.

#### Hypocotyl length

Hypocotyl length (Legee and Smith, 2002) of *G. hirsutum* varieties was improved at the Marrietta fine sandy loam location when grown in *R. reniformis* infested soils in

the presence of a nematicide with exception of Stv 5458 which did not differ between treated or untreated (Table 4.4). Across all varieties, treated Stv 5288 exhibited the greatest hypocotyl length compared to all other varieties, regardless of seed treatment, except treated Phy 499 or Stv 5458. Stv 5288 and Phy 499 had less hypocotyl growth with NST at the Leeper silty clay loam location while the other varieties were the same or improved with the NST.

## Influence of varieties grown in *R. reniformis* infested soils with and without nematicides during fruiting

Node of First Fruiting Branch

Initiation of node of first fruiting branch has been been used extensively to document treatment effects on harvest maturity (Jenkins et al., 1995). Nematicide treatments lowered initiation of NFFB in Phy 499 and Stv 5458 in Marrietta fine sandy loam soils (Table 4.4) and in Phy 375, Phy 499 and Stv 5458 at the Leeper silty clay loam soil location (Table 4.5). Later maturing varieties Stv 5458 and Phy 499 exhibited reduced NFFB in response to NST at both locations. Untreated and treated early maturing cotton varieties, Phy 375 and Stv 5288 grown at the Marrietta fine sandy loam location did not differ (Table 4.2). However, Phy 375, Phy 499 and Stv 5458 at the Leeper silty clay loam location initiated fruiting lower when a nematicide was applied (Table 4.5).

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Expression of varieties relative to plant height grown in *R. reniformis* infested soils with and without nematicides during fruiting

Square growth period

All varieties treated with NST had increased plant height during the square evaluation period at the Marrietta fine sandy loam location with exception of Phy 499 which did not differ between treated or untreated (Table 4.6). Phy 499 lack of growth differences between treated and untreated could be due to its being a late maturing variety. Stv 5288 had reduced node number with NST while Phy 499 had an increased node number with NST. There was no difference between NST and untreated in the remaining varieties (Table 4.6). The NST improved HNR for Stv 5288 and FM 1740, but did not influence the remaining treatments (Table 4.6).

At the Leeper silty clay loam location (Table 4.7), all varieties were taller as result of the exception of Phy 499, which did not differ from the untreated. At the Leeper silty clay loam location, there was no difference between variety or NST vs. untrerated on TN (Table 4.7). Height to node ratio was greatest in Phy 499 regardless of treatment, which was also the tallest variety (Table 4.7). Seed treatment did impact plant height for Phy 499 for either soil type, suggesting Phy 499 had some tolerance to *R. reniformis*.

#### Bloom growth period

Plant height of all treated varieties at bloom was improved at the Marrietta fine sandy loam location when NST was used (Table 4.6). Due to intrinsic growth patterns, treated FM 1740 and Stv 5288 did not differ in plant height from the untreated Stv 5458 or Phy 499. Height to node ratio increased with NST at bloom in all varieties, except for Stv 5458. Stv 5458 exhibited greater nematode tolerance than other varieties at the Marrietta fine sandy loam location since HNR was not impacted by NST. At the Leeper silty clay loam location, plant height at bloom increased in all varieties with the NST (Table 4.7). Similarly, HNR increased between untreated and NST in all varieties except Phy 375 which showed no difference in HNR, but did increase in TN. With the exception of Phy 375, nematicide treatments resulted in increasing node development at the Leeper silty clay loam location, and NST plants had longer internodes indicating seed treatment mitigated the impact of *R. reniformis* in all varieties except Phy 375 during bloom.

#### Open boll growth stage

Plant height at open boll at the Marrietta fine sandy loam location (Table 4.6) was increased by NST except for FM 1740 which is a naturally shorter variety. Stv 5458 with NST was the tallest variety but Stv 5458 also outperformed all other varieties when untreated. With exception of Stv 5458, all varieties had more total nodes with NST than when untreated (Table 4.6). Height to node ratio increased with NST in all varieties, except FM 1740 and Phy 499. Phy 499 increased in height but also had a large increase in node number with NST resulting in no difference in HNR. Similar results were measured at the Leeper silty clay loam location except Phy 499 had no difference in height between NST and untreated (Table 4.7). At the Leeper silty clay loam location, treated Phy 375, Stv 5288 and Phy 499 were similar in height. Total nodes decreased with NST in Phy 375 and Phy 499, increased with NST in FM 1740 and Stv 5458 and was similar to Stv 5288 (Table 4.7). However, HNR increased with NST for all varieties tested (Table 4.7). Varieties have differing tolerance in *R. renifromis* infested soils as

shown in non-nematicide treatments. However, location seemingly impacts how well varieties perform under *R. reniformis* environments.

Variety average plant height by node grown in *R. reniformis* infested soils Square growth phase

Evaluation of plant height by node provides a powerful method by which the variety performance relative to a stress effect can be quantified and indentified via internode elongation (Kerby et al., 2003). With this method, each internode is measured in cumulative fashion culminating with a total plant height. In such, effects of stress events can be measured by observing growth at each internode since during plant development three unexpanded nodes exist in meristematic tissues of the terminal at any point in time. Development of these nodes is greatly affected by growth conditions in the field. At the Marrietta fine sandy loam location (Table 4.8), there were no differences due to treatment within varieties at main-stem node one or three. At node one, Sty 5288 (NST and untreated) was taller than other varieties and treatments although treatment had no effect on growth of Stv 5288. Internode length at main-stem node five indicated no differences between treated and untreated for varieties Phy 375, Sty 5288 and Sty 5458. At node five, the NST treated FM 1740 and Phy 499 were taller than the untreated suggesting these varieties experienced stress from nematodes earlier than other varieties. Average plant height at main-stem node nine indicated that NST treatment was beneficial within all varieties except Phy 375. At main-stem node eleven, all varieties were taller than the untreated comparison when treated with an NST. This remained true through main-stem node thirteen except for Phy 499 where the height was similar for NST and untreated plants. By main-stem node fifteen, all varieties were taller when receiving a

NST. Phy 375 was as much as one inch taller when treated with a NST compared to only about one-half of an inch increase in other varieties. Lack of difference in height between NST and untreated Phy 375 up to main-stem node eleven suggests a higher tolerance to nematode populations in this variety at this growth stage.

Unlike at the Marrietta fine sandy loam location, Stv 5288 and FM 1740 at node one at the Leeper silty clay loam location (Table 4.9) were taller in NST than the untreated plants. Average plant height at main-stem node three showed no differences between the NST or untreated plants within variety. At main-stem node five and seven, only FM 1740 and Stv 5458 had increased plant height when treated with the NST. All other varieties were similar in the untreated or the NST. Average plant height at mainstem node nine followed a similar pattern as in the previous nodes, except Stv 5288 along with Stv 5458 and FM 1740 increased in plant height with the NST. This may indicate these varieties show early nematode tolerance but lose tolerance by node nine. Phy 499 treatments were shorter than other treatments. Average plant height at main-stem node eleven is where nematodes appear to have affected growth at the Leeper silty clay loam location with all varieties increasing in height with NST treatment. This is different than at the Marrietta fine sandy loam location where this break occurred at node nine for all varieties except Phy 375. This pattern held true through node 15. Nematicides improve growth in *R. reniformis* infested soils through square production, but the response is variety driven. Overall, effect of nematodes on plant height occured earlier at the Marrietta fine sandy loam location.

Average plant heights by node during bloom growth phase

Internode elongation of nodes one through five was considered to be complete at square growth stage. The second evaluation of internode growth was measured at bloom phase and included nodes eleven through 21. At the Marrietta fine sandy loam location, all varieties, except Stv 5458 saw growth advantages from the Aeris® treatments at mainstem node eleven during bloom (Table 4.10). All Aeris<sup>®</sup> treated varieties grown at the Leeper silty clay loam location at node eleven had taller growth compared to untreated (Table 4.11). At the Marrietta fine sandy loam location, all varieties were affected by nematodes except Stv 5458 which showed no height difference between treated and untreated from node 11 to 21. Growth of Stv 5458 at the Marrietta fine sandy loam location at bloom indicated tolerance to nematodes at later growth stages than when measured at square. At the Leeper silty clay loam location, all varieties benefitted from the NST from nodes 11 to 15, with the exception of Sty 5288 at node 15 (Table 4.11). In the Marrietta fine sandy loam location, Phy 499 was unaffected by NST. This was not observed at the Leeper silty clay loam location where Stv 5458 benefitted from the NST through node 21 measured at bloom. Phy 499 showed no height difference at the Leeper silty clay loam location due to NST from nodes 17 to 21 indicating tolerance to nematodes with plant maturity or cessation of growth after node 17. Phy 375 exhibited a 15% and 10% increase in plant height at node 21 for Marrietta fine sandy loam and Leeper silty clay loam locations, respectively. This indicates Phy 375 may have a lower R. reniformis tolerance of the varieties tested and suggests that Phy 375 requires maintenance in this environment (Usery et al., 2005; Legee et al., 2007, Blessitt et al., 2012).

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Percent sympodial fruit retention by position across all fruiting zones of cotton varieties grown in *R. reniformis* infested soils

Square fruiting period

Main stem fruit retention at fruiting position one indicated no difference in percent fruit retention between Aeris<sup>®</sup> and the untreated within variety at either location (Table 4.12 and 4.13). Fruit retention at sympodial fruiting position two in Marrietta fine sandy loam location was improved at square with Aeris<sup>®</sup> in Phy 375, Phy 499 and Stv 5458 (Table 4.12). At this fruiting position, Aeris<sup>®</sup> treatments improved retention as was seen in the previous nematicide study (i.e. Chapter 3). Fruit retention at square did not differ at sympodial position two between varieties when treated with Aeris<sup>®</sup> for the Leeper silty clay loam location (Table 4.13). Fruit retention at position two was greatest in Stv 5288, FM 1740 and Phy 499 indicating these varieties had improved retention farther out the fruiting branch, but still at a high quality position (Jenkins et al., 1990 a and b; Sadras, 1995).

At square, fruit retention at position >2 in Marrietta fine sandy loam soils was improved with NST treatment improved fruit retention for Stv 5288 and Stv 5458 (Table 4.12). Aeris<sup>®</sup> treated and untreated FM 1740 had greater retention than Aeris<sup>®</sup> treated and untreated Phy 375 and Phy 499 at fruiting position > 2. At the higher Leeper silty clay loam location, Aeris<sup>®</sup> treatment did not improve main-stem retention at fruiting position > 2 of Phy 375, Stv 5288 or Stv 5458 (Table 4.13). Phy 499 and FM 1740 had improved retention with addition of Aeris<sup>®</sup>. Improvement in fruit retention at square using Aeris<sup>®</sup> was dependent on variety at positions farther out the sympodial branch.

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Bloom growth phase

Aeris<sup>®</sup> improved fruiting position one retention during bloom (Table 4.12) in all varieties grown at the Marrietta fine sandy loam location with exception of Stv 5288 and FM 1740 which did not benefit from Aeris<sup>®</sup> treatment. At the Leeper silty clay loam location at fruiting position one, Phy 375, Phy 499, FM 1740 and Stv 5458 had improved fruit retention with addition of Aeris<sup>®</sup> (Table 4.13). When treated, FM 1740 and Phy 499 had the greatest fruit retention at fruiting position one. During this growth phase, repartitioning of carbohydrates for boll development was on-going (Schubert et al.1986; Sadras, 1995) and retention was greatly reduced especially in untreated varieties in response to *R. reniformis* populations (Cook et al., 1997b; Smith et al., 1996; Jones et al., 1959).

Fruit retention at bloom at fruiting position two within at the Marrietta fine sandy loam location (Table 4.12) indicated only Stv 5458 had increased fruit retention with Aeris<sup>®</sup> treatment. At the Leeper silty clay loam location, all Aeris<sup>®</sup> treated varieties had improved fruit retention at fruiting position two except Stv 5288 (Table 4.13). Untreated Stv 5288 had as great fruit retention as treated Phy 499. A higher fruit retention at fruiting position two indicates compensation for a lower retention level at fruiting position one (Jenkins et al., 1995; Sadras, 1995).

Fruit retention at position > 2 at bloom was increased with addition of Aeris<sup>®</sup> for all varieties at the Marrietta fine sandy loam location with exception of FM 1740 (Table 4.12). FM 1740 had lower fruit retention than all other treated varieties except untreated Phy 499, Phy 375 or Stv 5458. At the Leeper silty clay loam location (Table 4.13), similar results were observed as at the Marrietta fine sandy loam location in fruit retention at position > 2 at bloom. FM 1740 again had no difference in fruit retention between treated and untreated. FM 1740 had greater fruit retention than Aeris<sup>®</sup> treated or untreated Stv 5458 and Phy 375. In Leeper silty clay loam at this fruiting position, Stv 5288 had the greatest fruit retention of the varieties tested with application of Aeris<sup>®</sup>.

#### Open boll growth stage

In the Marrietta fine sandy loam location, fruit retention at position one measured at open boll was improved in varieties treated with Aeris<sup>®</sup> except FM 1740 (Table 4.12). FM 1740 did not benefit from the Aeris<sup>®</sup> and had lower fruit retention compared to other NST treatments. In addition, all varieties when untreated did not differ from the treated or untreated FM 1740. Varieties with greatest fruit retention were Aeris<sup>®</sup> treated Phy 499 and Stv 5288. At the Leeper silty clay loam location, all varieties increased fruit retention with Aeris<sup>®</sup> seed treatment (Table 4.13). Unlike at the Marrietta fine sandy loam location, FM 1740 treated had the greatest fruit retention at position one at open boll and exhibited tolerance to *R. reniformis* at the Leeper silty clay loam location.

Percent fruit retention at sympodial fruiting position two at the Marrietta fine sandy loam location measured at open boll followed the same pattern as at position one (Table 4.12). Only FM 1740 had no increase in fruit retention with Aeris<sup>®</sup>. Treated Phy 375 had greater fruit retention than the remaining treated varieties benefitting from the presence of the nematicide at the Marrietta fine sandy loam location. Treated FM 1740 had a greater fruit retention than untreated Phy 375, Stv 5288, Phy 499 and Stv 5458 (treated or untreated). Fruiting position two retention differences indicate *R. reniformis*  populations negatively impacted most varieties diminishing fruit retention at this fruiting position; however, FM 1740 showed excellent nematode tolerance at the Marrietta fine sandy loam location. This further demonstrates the impact of *R. reniformis* by cotton variety on fruit retention along the sympodial branch.

Percent fruit retention at position two of *G. hirsutum* varieties grown in a soil of Leeper silty clay loam infested with *R. reniformis* increased when Aeris<sup>®</sup> seed treatment was applied except for Stv 5288 (Table 4.13). Stv 5288 was comparable to treated Stv 5458, FM 1740 and untreated Phy 375 in fruit retention. Aeris<sup>®</sup> treated Phy 375 had the greatest fruit retention compared to all other tested varieties.

Position > 2 fruit retention at the Marrietta fine sandy loam location (Table 4.12) was similar within variety, regardless of treatment, except untreated Phy 375 had greater retention. Untreated Phy 375 had greater fruit retention compared to treated plants indicating variety sensitivity to *R. reniformis*. Delayed harvest maturity was due to lower fruit retention as untreated Phy 375 had the greatest fruit retention during open boll at position > 2 followed by Stv 5288.

At the Leeper silty clay loam location (Table 4.13), all Aeris<sup>®</sup> treatments resulted in lower retention at fruiting position > 2 except FM 1740 which had improved fruit retention at open boll. This is driven by a lower fruit retention of earlier fruiting sites as seen in the untreated varieties (Jenkins et al., 1995; Smith et al., 1996; Sadras, 1995). At this location, all untreated varieties differed from each other except Phy 375 and Stv 5458. Of all the treated varieties, FM 1740 had the least fruit retention when untreated and the most fruit retention when treated with Aeris<sup>®</sup>.

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During this evaluation period, first and second position fruit retention provided a greater understanding how individual varieties respond to the presence of *R. renifromis* across soils. Retention at the >2 fruiting site results in delayed harvest maturity resulting from fruit loss at earlier fruiting positions. All Aeris<sup>®</sup> treated varieties, except FM 1740, were improved in retention at the high quality positions, one and two, when compared to untreated varieties. However, higher retention at these fruiting sites established early during square and then bloom in untreated varieties indicate variety tolerance to *R. reniformis* resulting in a higher degree of retention between bloom and open boll growth stages.

Untreated varieties did provide benefit at position one, despite the location, through bloom but were reduced greatly between bloom and open boll which followed the increase in *R. reniformis* population (Tables 4.1, 4.2 and Fig. 4.1).

# Cotton maturity measured as nodes above white flower, nodes above cracked boll, percent open boll

Nodes Above White Flower across two locations

Number of nodes above white flower (NAWF) is a measure of harvest maturity in cotton. At the Marrietta fine sandy loam location (Table 4.14), NAWF was reduced in all varieties when treated with Aeris<sup>®</sup> except Stv 5288 and Phy 499 due to increased boll retention and partitioning of carbohydrates into a higher number of bolls (Jenkins et al., 1995; Sadras, 1995). Untreated Phy 375 and Stv 5288 did not differ in NAWF from Aeris<sup>®</sup> treated FM 1740, a later maturing variety. Few differences in NAWF were found in the Leeper silty clay loam location (Table 4.15), and only Phy 499 had a decrease in NAWF with the Aeris<sup>®</sup> indicating an improvement in harvest maturity. A lack of NAWF

differences due to Aeris<sup>®</sup> treatment during bloom indicates nematodes had little effect on plant maturity during this growth phase further indicating a level of variety tolerance.

Nodes Above Cracked Boll and Percent Open Boll

Nodes above cracked boll and percent open boll are development criteria highly indicative of maturity within a treatment or variety and is greatly enhanced by increased boll numbers (Jenkins et al., 1995). All Aeris<sup>®</sup> treated varieties possessed lower NACB at the Marrietta fine sandy loam location except with FM 1740 (Table 4.14). FM 1740 had greater tolerance to *R. reniformis* in NACB having no differences between Aeris<sup>®</sup> treated and untreated plants. Percent open boll was increased in all varieties when treated with Aeris<sup>®</sup>. While FM 1740 did not differ in NACB with addition of Aeris<sup>®</sup>, there was an increase in percent open boll. Node above cracked boll and percent open boll had a direct correlation with total fruit retention which provided a sink for carbohydrate partitioning (Schubert et al., 1986, Sadras, 1995).

Node above cracked boll and percent open boll of all varieties grown at theLeeper silty clay loam location (Table 4.15) were improved with the Aeris<sup>®</sup> treatment; NACB were reduced and open boll increased. Untreated Phy 375 had higher NACB than other untreated varieties indicating this variety is very sensitive to *R. reniformis*. Aeris<sup>®</sup> treated varieties did not differ from each other except for FM 1740 which which had greater NACB than the remaining treated varieties.

Production of monopodial branches and bolls of *G. hirsutum* among treated and untreated varieties grown in *R. reniformis* infested soils

Monopodial branch production

Monopodial branch and subsequent boll production occurs after initiation of sympodial branches and is increased under lower plant populations, wider planting rows and vigorous growing conditions (Mauney, 1986). At both locations (Table 4.16), all varieties had greater monopodial branch number when treated with Aeris<sup>®</sup> except FM 1740 at both locations and Stv 5458 at the Leeper silty clay loam location. Nematicide treatment did improve growth and overall plant health of some varieties like Phy 375, Stv 5288 and Phy 499 when compared to untreated plants as indicated by increased monopodial branch numbers.

## Monopodial boll production

Monopodial boll production per plant at the Marrietta fine sandy loam location (Table 4.16) exhibited fewer differences than did as monopodial branch production. Aeris<sup>®</sup> treated varieties, Stv 5458, Stv 5288, and FM 1740 did not differ from untreated plants. Growth continued in absence of a nematicide suggesting some *R. reniformis* tolerance.

At the Leeper silty clay loam location (Table 4.16), monopodial boll production was lower in untreated Phy 375, Phy 499, and Stv 5458 compared to treated in each variety. Nematicide seed treatment did not increase monopodial boll production for Stv 5288 or FM 1740 indicating tolerance. Phy 375 treated with Aeris<sup>®</sup> had 2.4 more monopodial bolls compared to untreated Phy 375 plants. Stalk and boll diameters of G. hirsutum grown in *R. reniformis* infested soils Basal stalk diameters in *R. reniformis* infested soils

Basal stalk size becomes important to *G. hirsutum* due to its excessive biomass generated during boll development and need to reduce lodging. At the Marrietta fine sandy loam locations (Table 4.17), Aeris<sup>®</sup> treated Phy 375, Stv 5288, and Stv 5458 had thicker basal stalks than untreated. Aeris<sup>®</sup> treated Stv 5458 had a larger basal stalk diameter than the other treated varieties. Phy 499 and FM 1740 did not differ when treated with Aeris<sup>®</sup>, indicating nematode tolerance at the Marrietta fine sandy loam location. Basal stalk diameter at the Leeper silty clay loam location (Table 4.17) was larger in Aeris<sup>®</sup> treated Stv 5458, Phy 499 and Stv 5288. As at the Marrietta fine sandy loam location, there was evidence of variety tolerance to *R. reniformis*.

Variety effects on boll diameters in *R. reniformis* infested soils at two locations

Boll diameter measurement at 12 nodes below the terminal at the Marrietta fine sandy loam location was increased for Phy 375 and Stv 5288 treated with Aeris<sup>®</sup>. Boll diameter at 12 nodes below the terminal (represents older bolls) at the Leeper silty clay loam location (Table 4.17) increased boll size in all varieties except FM 1740 when treated with Aeris<sup>®</sup> seed treatment. Aeris<sup>®</sup> treated Phy 499 produced larger bolls than any other variety and treatment combination. FM 1740 treated with Aeris<sup>®</sup> did not differ from remaining treated varieties except Phy 499 which produced larger bolls and Stv 5288 which produced smaller bolls. This indicates FM 1740 has some tolerance in *R. reniformis* infested soils. Untreated Phy 375, Stv 5458 and Stv 5288 had reduced boll size at this node.

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Boll diameter measurements at nine nodes (represents younger boll) below the terminal indicated improved boll growth from Aeris<sup>®</sup> seed treatment with exception of Stv 5288 at the Marrietta fine sandy loam location and Phy 375 at the Leeper silty clay loam location. At the Marrietta fine sandy loam location boll diameter difference for all Aeris<sup>®</sup> treated varieties differed and were as follows: Phy 375, Phy 499, Stv 5458, FM 1740 and Stv 5288. With the exception of Stv 5288, untreated varieties had higher boll diameter differences when compared within treated varieties.

Effects on boll diameter at nine nodes below the terminal by variety at the Leeper silty clay loam location indicated all varieties had increased boll diameter with Aeris<sup>®</sup> treatment except Phy 375 which had no difference (Table 4.16).

Differences between node nine and node 12 from the terminal indicate a difference in boll rate of development between these nodes. Bolls at 12 nodes below the terminal generally had enough time to allow boll size to equalize. However, at nine nodes below the terminal, bolls were still developing facilitating measurement of developmental delays. At the Marrietta fine sandy loam location (Table 4.17), all untreated varieties had greater boll diameter differences than treated except Stv 5288 which exhibited no treatment effect. This indicates nematicides do hasten boll development. Boll diameter difference was 75% greater in in Phy 375 suggesting sensitivity to *R. reniformis*. Phy 375, Stv 5288, FM 1740 and Stv 5458 had increases in boll diameter differences when untreated compared to treated at the Leeper silty clay loam location (Table 4.17).

Yield of treated and untreated upland cotton varieties grown in *R. reniformis* infested soils

Yield was increased at the Marrietta fine sandy loam location with addition of Aeris<sup>®</sup> with exception of FM 1740 and Stv 5458 (Table 4.18). Phy 375 had greatest yield, compared to all other treatments, and yield increase between treated and untreated equaled 430 lbs lint/Ac. Aeris<sup>®</sup> treated Stv 5288 and Phy 499 did not differ from each other and except treated Phy 375, were greater in yield than the remaining treatments. Aeris<sup>®</sup> treated FM 1740 did not differ from treated Stv 5458 or untreated Stv 5288. All varieties except Phy 375 yielded similarly without the nematicide.

At the Leeper silty clay loam location (Table 4.18), Phy 375, Stv 5288 and Stv 5458 had greater yield when treated with Aeris<sup>®</sup>. Phy 499 regardless of Aeris<sup>®</sup> treatment performed better compared to all other treatments except treated Stv 5458. Although Aeris<sup>®</sup> treated Phy 499 yielded more than Stv 5458, untreated Phy 499 was similar to both. Phy 375 yielded less than all other treatments. Treatment with Aeris<sup>®</sup> increased lbs lint/Ac by 173 lbs and 167 lbs for Phy 375 and Phy 499, respectively. Stv 5288 treated with Aeris<sup>®</sup> was able to produce greater yields than Phy 375 at both locations, which was supported by previous growth parameters. Phy 499 and FM 1740 show moderate *R. reniformis* tolerance and yielded well at the Leeper silty clay loam location. This data agrees with previous findings where some commercial varieties showed a level of tolerance to *R. reniformis* (Blessit et al. 2012; Legee et al., 2007), but nematicides are still beneficial in these environments (Phipps and Eisenback, 2005).

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# Growth of Aeris<sup>®</sup> treated and untreated cotton varieties under varying populations of *R. reniformis* in a greenhouse environment

Effects of R. reniformis population on root biomass development

Within the greenhouse environment, all Aeris<sup>®</sup> treated varieties had increased root biomass at all nematode populations including zero (Table 4.19). In absence of R. reniformis. Aeris<sup>®</sup> treated varieties developed larger root volumes compared to untreated plants. This establishes root growth parameters in a stress free environment. Varieties in absence of *R. reniformis* demonstrating the greatest root volumes included Phy 375 and Phy 499 which did not differ from each other. One variety, untreated Phy 499, displayed no reduction in root growth despite R. reniformis population with exception of Pi 7,500. Even though root growth declined in treated Phy 499 as *R. reniformis* population increased, it displayed greater root biomass than other treatments at both Pi 5,000 and 7,500 showing some *R. reniformis* tolerance at higher populations. In absence of a nematicide, Phy 499 root development was reduced at Pi 7,500 to levels similar to other varieties. Remaining varieties treated with Aeris® indicated increased root biomass. As population increased to Pi 5,000, root growth differences among varieties treated with Aeris<sup>®</sup> began differing from each other with Stv 5458 having greater root volume than Phy 375. At Pi 5,000, treated FM 1740 and Sty 5288 root volumes did not differ from each other, but had lower volume than Stv 5458 and Phy 499. As R. reniformis population increased to Pi 7,500, only Aeris<sup>®</sup> treated Phy 499 and FM 1740 produced greater root volumes compared to Phy 375, Stv 5288, and Stv 5458, regardless of treatment. As *R. reniformis Pi* increased to 5,000, untreated Phy 375 and Stv 5288 produced less root volume compared to all other treatment combinations. As R. reniformis population increased to Pi 7,500, each untreated variety produced less root

mass compared to all other treatment combinations and populations except untreated Phy 375 at *Pi* 5000. This data indicates Aeris<sup>®</sup> adds to tolerance of *R. reniformis*, but root biomass reduction occurs with use of Aeris<sup>®</sup> as *R. reniformis* population increases. Aeris<sup>®</sup> efficacy was reduced in all varieties at *Pi* 7,500. In addition, there was some variety tolerance to *R. reniformis* until the population reached *Pi* 7,500.

Effects of R. reniformis population on shoot biomass development

Shoot biomass in the greenhouse environment followed a similar pattern as root biomass among varieties with Aeris<sup>®</sup> treatment improving shoot biomass with the exception of Phy 375 when Pi is 0 or 5,000 (Table 4.19). For varieties other than Phy 375, this establishes genetically determined shoot development parameters and indicates nematicide, even in environments void of R. reniformis, improves shoot growth and development. Aeris<sup>®</sup> treated Stv 5288, Phy 499 and FM 1740 void of *R. reniformis* had greater shoot biomass than Phy 375. Aeris® treated Stv 5458 did not differ from FM 1740 or Phy 375. Shoot biomass production across Aeris<sup>®</sup> treated varieties declined at varying degrees depending on variety as *R. reniformis* population increased. Aeris<sup>®</sup> treated Phy 375 had less shoot biomass compared to other Aeris<sup>®</sup> treated varieties except FM 1740 at Pi 2,500. At Pi 2,500, Aeris<sup>®</sup> treated Stv 5288, Phy 499 and Stv 5458 had greater shoot biomass than Phy 375 while not differing from each other. Further separation continued at the R. reniformis Pi 5,000 with Aeris® treated Stv 5458 and Phy 499 differing from Phy 375 FM 1740, and Stv 5288 at this population. Within the highest R. reniformis Pi 7,500, Aeris<sup>®</sup> treated Phy 499 maintained greater shoot biomass production over the remaining varieties. This indicates Phy 499 is more tolerant to nematode populations or shoot biomass responds better to nematicide.

Recovered R. reniformis juvenile numbers by variety

*Rotylenchulus reniformis* is an obligate parasite and requires developing root mass for reproduction (Linford and Oliveira, 1942). A direct correlation of root biomass and increased population of *R. reniformis* has been observed in previous work (Lawrence and McLean, 1995; Lawrence and McLean, 1996). It is fair to say that higher juvenile numbers also relates to increased root biomass as was observed (Tables 4.19 and 4.20).

As with root growth and development, Aeris<sup>®</sup> treated Phy 499 had the highest number of *R. reniformis* juveniles recovered regardless of Aeris<sup>®</sup> treatment or *Pi*. This was also observed in the untreated Phy 499 at *Pi* 5,000. Aeris<sup>®</sup> treated Phy 375 had the lowest recovered nematodes as *R. reniformis* population increased to *Pi* 5,000. Within Aeris<sup>®</sup> treated varieties at *Pi* 2,500, juvenile numbers recovered were higher in Phy 499 suggesting Aeris<sup>®</sup> increased tolerance to *R. reniformis*. Possibly, this is due to Phy 499's indeterminate nature possessing a slow developing root system. Untreated Phy 499, Stv 5288, and Stv 5458 had the highest numbers of juveniles recovered compared to Phy 375 or FM 1740 at *Pi* 2,500. Treated and untreated Stv 5458 and Stv 5288 had no differences within varieties in juveniles recovered indicating these varieties at this population benefited little from the nematicide treatment.

A decline in juvenile numbers recovered occurred at *Pi* 5,000 without NST use, regardless of variety. This supports previous research by Lawrence & McLain (1996) indicating greater nematode juvenile populations occur at higher root biomass. Treated Stv 5458 had more juveniles recovered indicating root mass probably benefited from the nematicide. At *Pi* 5,000, Stv 5288 juvenile recovery declined due to restricted root development compared to lower *Pi* resulting from increased feeding. Treated FM 1740,

despite being different from treated Stv 5458, declined as *R. reniformis* population increased.

All varieties benefitted from Aeris<sup>®</sup> treatment at *Pi* 7,500. Within this population, treated or untreated FM 1740 were comparable to treated or untreated Phy 499 in juveniles recovered while treated Stv 5288 and Stv 5458 did not differ from Phy 375.

## Recovered R. reniformis egg numbers by variety

The same pattern occurring in juveniles was observed in egg production indicating reproduction was reduced as root biomass declined (Tables 4.19 and 4.20). Evaluation of untreated varieties for egg recovery across varieties show-cases reproduction of *R. reniformis* effects by variety. Under low nematode populations, root growth of Phy 375 was reduced and complete reproduction of R. reniformis declined further indicating sensitivity to R. reniformis. Untreated FM 1740 and Stv 5288 followed in this reproduction pattern, but did differ from each other and had more eggs recovered than Phy 375. At this population Stv 5288 had greater egg recovery than FM 1740. Untreated Phy 499 and Stv 5458 had greatest egg recovery indicating greater reproduction. As population increased to Pi 5,000, Phy 375 had continued low egg recovery indicating further increased sensitivity to *R. reniformis*. Remaining untreated varieties continued supporting nematode reproduction as they did not differ from each other and had greater egg recovery than Phy 375. The greatest egg recovery at Pi 7,500 occurred in treated Phy 499, Stv 5458 and FM 1740 varieties. Varieties had different tolerances to *R. reniformis* with tolerance enhanced with use of a nematicide. This synergistic relationship can be used to improve crop yield and improve profitability.

Effect of *R. reniformis* population on growth of *G. hirsutum* varieties Effect of *R. reniformis* on plant height

In absence of *R. reniformis*, plant height was greatest in Phy 499 and Stv 5458 compared to FM 1740, a genetically shorter variety (Table 4.21). Stv 5288 did not differ from Phy 375. Comparison of Aeris<sup>®</sup> treated plants to untreated indicated increased height even in absence of *R. reniformis*. As *R. reniformis* population increased, plant height decreased. Plant height of Aeris<sup>®</sup> treated plants at *Pi* 2,500 was greater than the untreated for all varieties. Aeris<sup>®</sup> treated Phy 499, Stv 5458 and Stv 5288 were taller than FM 1740 and Phy 375 but did not differ from each other. Untreated Phy 375 was shorter than all other untreated varieties at *Pi* 2,500, except FM 1740. As the inoculated treatment population increased to 5,000 *R. reniformis*, a similar pattern of height was observed to that observed at *Pi* 2,500 with Aeris<sup>®</sup> treated Phy 499 and Stv 5458 at *Pi* 5,000 population did not differ from each other, but did display greater plant height compared to Phy 375 and Stv 5288. At *Pi* 7,500, untreated Phy 499 and Stv 5458 were similar in height and were taller than Phy 375 and Stv 5288.

Treated and untreated FM 1740 had the smallest change in plant height as *R*. *reniformis* populations increased indicating its ability to not deviate greatly from its genetically governed plant height in the presence of *R. reniformis*. In addition, Aeris<sup>®</sup> treated Stv 5288 did not differ in plant height from FM 1740 at *Pi* 7,500. All varieties were reduced in plant height at *Pi* 7,500 compared to *Pi* 2,500 and did benefit from the nematicide application. Interestingly, treated FM 1740 had no difference when compared to untreated while Stv 5288 and Phy 499 were different in height. Stv 5288 is tolerant to low to moderate *R. reniformis* populations while Phy 375 is intolerant at all populations.

### Effect of R. reniformis population on total node production

In absence of *R. reniformis*, Aeris<sup>®</sup> treated plants had greater total node production compared to untreated plants for all varieties except Phy 499 (Table 4.21). Total node production was reduced compared to treated as *Pi* increased for all varieties when untreated except Phy 499. Node number for all Aeris<sup>®</sup> treated varieties at *Pi* 2,500 were the same as with no nematodes. While total node number at *Pi* 5,000 remained the same for Phy 499, all other Aeris<sup>®</sup> treated varieties had fewer nodes than at *Pi* 0. As with plant height, Phy 499 had the greatest tolerance to nematode populations. At the highest nematode population there was no difference in total node number between treated Phy 499 and Stv 5458. Aeris<sup>®</sup> treatment improved total node production compared to untreated plants, and variety tolerance was observed in absence of nematicides.

Effect of *R. reniformis* population on HNR (Height to Node Ratio)

Stv 5288 HNR increased when treated with Aeris<sup>®</sup> at *Pi* 0 and *Pi* 5,000 while Phy 375 only increased with treatment at *Pi* 5,000. All Aeris<sup>®</sup> treated varieties had greater HNR than Phy 375 at *Pi* 2,500. At *Pi* 5,000, Aeris<sup>®</sup> treated Phy 375 had lower HNR than other varieties. Aeris<sup>®</sup> treated varieties Stv 5288 and Phy 375 had lower HNRs at *Pi* 7,500 compared to Phy 499. Phy 375 intolerance to *R. reniformis* was further demonstrated and began losing tolerance at *Pi* 2,500. Another variety possessing low to moderate tolerance was Stv 5288 which declined in HNR at *Pi* 5,000. Phy 499, Stv 5458

and FM 1740 continued to show tolerance in *R. reniformis* infested soils beyond *Pi* 5,000 based on HNR.

Effect of *R. reniformis* population on NFFB (Node of First Fruiting Branch)

Node of first fruiting branch (NFFB) is a good indicator of initiation of cotton harvest maturity. Higher NFFB above what is genetically governed is an expression of early stress that can lead to higher fruit initiation on the main axis and encourage lateness in *G. hirsutum* development delaying harvest maturity (Mauney, 1986). However, since there is a strong genetic influence, response at *Pi* 7,500 compared to *Pi* 0 only increased for FM 1740 regardless of Aeris<sup>®</sup> treatment or Phy 375 and Stv 5288 when untreated. However, severity of delayed fruiting was greatly accentuated where a nematicide was not used especially at higher *R. ren*iformis populations (Table 4.22).

In summary, initiation of fruiting is hastened with higher *R. reniformis* populations when a nematicide is used demonstrating value of using a nematicide in early growth and development of *G. hirsutum*. However, nematode tolerance as measured by fruit initiation is being exhibited within a variety as observed in untreated plants. Longer season varieties, Stv 5458 and Phy 499 tolerated nematode populations until *Pi* 7,500.

Effect of *R. reniformis* population on basal stalk development

Basal stalk diameter becomes important as *G. hirsutum* incurs heavy boll load and is predisposed to lodging prior to harvest (Mauney, 1986). Aeris<sup>®</sup> treatment increased basal stalk diameter in all varieties compared to untreated plants. This further supports plant health benefits from nematicide seed treatment. Within Aeris<sup>®</sup> treated plants in absence of *R. reniformis*, the only varieties with reduced basal diameter were Phy 375 and FM 1740 compared to Phy 499, Stv 5288, or Stv 5458 (Table 4.22). Basal stalk diameter remained larger at the *Pi* 2,500 in Stv 5288 compared to all other Aeris<sup>®</sup> treated varieties except Phy 499. Within this population, all varieties were increased in basal diameter when compared to untreated plants with exception of Stv 5458. No difference between varieties were observed between Aeris<sup>®</sup> treated plants at *Pi* 5,000 and 7,500. To determine strengths by variety in *R. reniformis* infested soils relative to basal stalk diameter, evaluation of untreated varieties across populations was conducted. Basal stalk development of Aeris<sup>®</sup> treated Stv 5288 was greater at *Pi* 2,500 compared to other varieties while there were no differences between Stv 5458, Phy 499 or FM 1740. Untreated Phy 375 produced smaller basal stalk diameters at *Pi* 2,500 and 5,000, indicating its lack of tolerance to *R. reniformis*. Basal stalk development at *Pi* 7,500 was greater in untreated Stv 5458 and Phy 499 compared to the remaining varietietis.

#### Conclusion

Commercially available varieties have some tolerance to *R. reniformis*. Understanding their growth characteristics allows for proper variety selection and mangement. Nematicide treatments did improve growth and yield of varieties in presence of *R. reniformis*. From these findings, Phy 375 was sensitive to *R. reniformis*, with improved growth when treated with a nematicide. Greenhouse studies further verified this variety's intolerance to *R. reniformis* as it began root biomass loss at *Pi* of 2,500. Stv 5288 followed a similar pattern, but was not affected as severely as Phy 375. In the greenhouse, Stv 5288 root biomass began deminishing at *Pi* 5,000. However, in the field, this variety in an untreated state out-yielded Phy 375 and showed smaller yield differences. From in the field studies, Phy 499 provided moderate tolerance to *R*. reniformis, but did have positive performance in greenhouse study as R. reniformis populations began to increase. A primary characteristic of Phy 499 that could have negatively impacted field performace was its natural late maturity making it difficult to manage in small plot environments. Phy 499 did perform well across both locations indicating its possible use across soils. Stv 5458 F and FM 1740 showed excellent performance in the field in presence of R. reniformis populations as well as under greenhouse environments. Sty 5458 performed well across both locations with the least differences between Aeris<sup>®</sup> treated and untreated varieties indicating its tolerance of *R*. reniformis. FM 1740 also demonstrated good performance in R. reniformis infested soils in the field and in the greenhouse. This variety had greatest yield at the Leeper silty clay loam location, but lower differences between Aeris® treated and untreated at the Marrietta fine sandy loam location tolerance in *R. reniformis*. Ranking of performance from most R. reniformis tolerant to least tolerant are as follows; Phy 499, FM 1740, Stv 5458, Stv 5288 and Phy 375. Those commercially available varieties showing the greatest tolerance are later maturing varieties having lower initial root growth. Under low populations of *R. reniformis*, these varieties could successfully produce adequate yield with only a nematicide.

In conclusion most varieties benefited from the presence of a nematicide (Phipps and Eisenback, 2005) but at varying degrees and performance can be improved by selecting the correct variety for the appropriate environment (Legee et al., 2007; Blessitt et al., 2012).

Table 4.1	In the field treatments of five commercial <i>G. hirsutum</i> varieties (Phy 375
	WRF, Phy 499 WRF, Stv 5458 B2RF, Stv 5288 B2RF, FM 1740 B2RF)
	treated and untreated with Aeris® seed treatment in R. reniformis infested
	soils.

Variety	Treatment <sup>z</sup>	Variety Maturity
	Aeris <sup>®</sup> @ 0.075 mg ai/seed rate	
PHY 375 WRF <sup>y</sup>	Untreated	Early
	Aeris <sup>®</sup> @ 0.075 mg ai/seed rate	
PHY 499 WRF <sup>y</sup>	Untreated	Full
	Aeris <sup>®</sup> @ 0.075 mg ai/seed rate	
STV 5458 B2RF <sup>x</sup>	Untreated	Mid
	Aeris <sup>®</sup> @ 0.075 mg ai/seed rate	
STV 5288 B2RF <sup>x</sup>	Untreated	Early
	Aeris <sup>®</sup> @ 0.075 mg ai/seed rate	
FM 1740 B2RF <sup>x</sup>	Untreated	Mid

<sup>z</sup> Aeris<sup>®</sup> was applied to the seed prior to planting by Bayer Crop Science (Raleigh, North Carolina) <sup>y</sup> Variety derivations: Phytogen (Phy) a subsidiary of Dupont. <sup>x</sup> Stoneville (Stv) and Fibermax (FM) subsidiaries of Bayer Crop Sciences (Raleigh,

North Carolina).

Table 4.2Five commercial G. hirsutum varieties (Phy 375 WRF, Phy 499 WRF, Stv5458 B2RF, Stv 5288 B2RF, FM 1740 B2RF) treated and untreated with<br/>Aeris® grown in varying populations of R. reniformis under greenhouse<br/>environment

Variety	Initial Nematode Population	Treatment	
PHY 375 WRF <sup>z</sup>	0		
	2,500	$\mathbf{A} : \mathbb{R} \bigcirc 0.075$ : (1)	Lintucato
	5,000	Aeris <sup>®</sup> @ 0.075 mg ai/seed rate	Untreate
	7,500		
PHY 499 WRF	0		
	2,500	$\mathbf{R} \subset \mathbf{R} \subset \mathbf{R}$	TTuturete
	5,000	Aeris <sup>®</sup> @ 0.075 mg ai/seed rate	Untreate
	7,500		
STV 5458 B2RF <sup>y</sup>	0		
	2,500	$\mathbf{A} : \mathbb{R} \bigcirc 0.075$ : (1)	Lintucato
	5,000	Aeris <sup>®</sup> @ 0.075 mg ai/seed rate	Untreate
	7,500		
STV 5288 BwRF	0		
	2,500	$\mathbf{A} : \mathbb{R} \bigcirc 0.075$ : (1)	Lintucato
	5,000	Aeris <sup>®</sup> @ 0.075 mg ai/seed rate	Untreate
	7,500		
FM 1740 B2RF	0		
	2,500	$\mathbf{A} : \mathbb{R} \bigcirc 0.075$ : (1)	Linturate
	5,000	Aeris <sup>®</sup> @ 0.075 mg ai/seed rate	Untreate
	7,500		

. Nematodes were pipetted in graduated fashion to autoclaved soils.

<sup>z</sup> Variety derivations: Phytogen (Phy) a subsidiary of Dupont. Stoneville (Stv) and Fibermax (FM) subsidiaries of Bayer Crop Sciences (Raleigh, North Carolina).

<sup>y</sup> Aeris<sup>®</sup> was applied to the seed prior to planting by Bayer Crop Science (Raleigh, North Carolina).

	М	arrietta Fin	e Sandy Lo		Leeper Fin	e Sandy Cl	ay	
	Renifor	m Nematod	le Numbers	/500cc <sup>v,w</sup>	Reniform Nematode Numbers/500cc <sup>v,w</sup>			
Treatment	May	June	July	Sept	May	June	July	Sept
	$(0 \text{ DAE}^{x})$	(40 DAE)	(70 DAE)	(100 DAE)	(0 DAE)	(40 DAE)	(70 DAE)	(100 DAE)
Phy 375 UT <sup>y</sup>	516.0b <sup>z</sup>	516.0c	1548.0d	26402.0ab	481.6ab	849.4cd	4289.2de	11029.6c
Phy 375 Trt <sup>y</sup>	1032.0ab	2365.0a	13351.6a	46762.4a	447.2ab	505.2de	9108.0c	21575.2a
Stv 5288 UT	580.6b	548.4c	1150.2d	11813.4b	825.6ab	1338.6b	2808.6ef	6493.0d
Stv 5288 Trt	680.6b	1580.2b	4912.8c	36786.6ab	481.6ab	559.0de	5074.0de	9546.0c
FM 1740 UT	516.0b	516.0c	2217.2d	16899.0b	1032.0a	2074.8a	1298.0f	5676.0d
FM 1740 Trt	548.3b	516.0c	559.0d	17834.2b	619.2ab	1032.0c	5504.0d	7256.2d
Phy 499 UT	548.2b	516.0c	1368.0d	29081.8ab	756.8ab	1419.0b	4450.6de	3751.8e
Phy 499 Trt	516.0b	1967.2ab	8127.0b	37377.8ab	412.8b	591.2de	13590.8a	22462.2a
Stv 5458 UT	516.0b	516.0c	1548.0d	26402.0ab	481.6ab	849.4cd	4289.2de	11029.6c
Stv 5458 Trt	1032.0ab	2365.0a	13351.6a	46762.4a	447.2ab	505.2de	9108.0c	21575.2a
LSD(0.05)	359.6	637.1	1726.2	17057.3	368.1	249.5	1893.4	1756.2

Table 4.3Seasonal population progression of *R. reniformis* across *G. hirsutum*<br/>varieties and soil types sampled at six cores per plot during four growth<br/>stages at Mississippi State University.

<sup>y</sup> UT=Untreated; Trt=Aeris<sup>®</sup>.

<sup>x</sup> DAE=Days After Emergence.

<sup>w</sup> Samples taken at rate of six per plot using a fluted probe from side of planted row.

<sup>v</sup> Samples taken at depth of 4.0 inches.

Table 4.4Influence of variety treated with Aeris® nematicide (Trt) and untreated (UT)in *R. reniformis* infested soils on plant population, node of first fruitingbranch, vigor and hypocotyl length (14 days after emergence) grown at theMarrietta fine sandy loam location.

Treatment	Plants/acre v	Node of First Fruiting Branch	Vigor (1-5) <sup>x</sup>	Hypocotyl Length (inches) <sup>y</sup>
Phy 375 UT	44,979.1b <sup>z</sup>	6.8c <sup>z</sup>	1.4bc	3.8c
Phy 375 Trt	42,782.8bc	6.6c	1.0c	4.3ab
Stv 5288 UT	44,230.4b	6.4c	1.4bc	4.1b
Stv 5288 Trt	43,781.3b	6.2c	1.0c	4.5a
FM 1740 UT	43,944.5b	6.4c	1.5b	3.7c
FM 1740 Trt	41,657.4c	6.2c	1.3bc	4.1b
Phy 499 UT	45,088.1b	8.8a	2.3a	3.7c
Phy 499 Trt	44,434.6b	7.9b	1.3bc	4.2ab
Stv 5458 UT	47,266.2a	7.9b	1.1bc	4.2ab
Stv 5458 Trt	47,211.8a	6.8c	1.1bc	4.2ab
LSD(0.05)	1799.2	0.8	0.3	0.2

<sup>z</sup> Means within columns followed by the same letter are not different according to the Least Significant Difference means separation test  $P\alpha$ =0.05.

<sup>y</sup> Hypocotyl measured from point of seed attachment to cotyledon.

<sup>x</sup> Visual vigor scale 1-5 where 1 had larger leaves and was taller while 5 was stunted.

<sup>v</sup> Plants/ac was conducted by counting all plants per plot with expanded cotyledons.

Table 4.5Influence of variety treated with Aeris® nematicide (Trt) or untreated (UT)in *R. reniformis* soils on plant population, node of first fruiting branch, vigor<br/>and hypocotyl length (14 days after emergence) grown at Leeper silty clay<br/>loam location.

Treatment	Plants/ac v	Node of First Fruiting Branch	Vigor (1-5) <sup>x</sup>	Hypocotyl Length (inches) y
Phy 375 UT	32,345.8d <sup>z</sup>	8.30b	3.10a	3.70b
Phy 375 Trt	29,822.7ef	7.40c	1.20cd	4.40a
Stv 5288 UT	31,184.1de	7.50c	3.00a	4.60a
Stv 5288 Trt	28,207.3f	7.40c	1.20cd	3.80b
FM 1740 UT	29,133.0ef	6.40d	2.90a	3.70b
FM 1740 Trt	28,370.6f	6.40d	1.40c	4.30a
Phy 499 UT	41,330.7a	10.0a	2.20b	4.50a
Phy 499 Trt	34,905.1c	8.40b	1.40c	3.70b
Stv 5458 UT	38,535.4b	10.1a	1.10cd	4.20a
Stv 5458 Trt	38,335.7b	8.5b	1.00d	4.40a
LSD(0.05)	1972.5	0.7	0.3	0.3

<sup>z</sup> Means within columns followed by the same letter are not different according to the Least Significant Difference means separation test  $P\alpha=0.05$ .

<sup>y</sup> Hypocotyl measured from point of seed attachment to cotyledon at 14 DAE.

<sup>x</sup> Visual vigor evaluation in ranking of 1-5 where 1 was best and 5 was worst at 14 DAE.

<sup>v</sup> Plants/ac was conducted by counting all plants per plot.

Table 4.6Gossypium hirsutum variety growth regarding growth parameters, total<br/>nodes, plant height, height to node ratio during square, bloom and open boll,<br/>in *R. reniformis* infested soils at the Marrietta fine sandy loam location.

	Square Bloom							Open Bol	1
Treatment		(40 DAE <sup>w</sup>	')	(7	70 DAE <sup>w</sup> )		(	(100 DAE <sup>v</sup>	v)
		Plant Ht <sup>x</sup>	HNR <sup>x</sup>		Plant Ht	HNR		Plant Ht	HNR
	TN <sup>x</sup>	(inches)	(inches)	TN	(inches)	(inches)	TN	(inches)	(inches)
Phy 375 UT <sup>y</sup>	14.0b <sup>z</sup>	14.9b	1.1b	20.8ab	28.6e	1.4e	19.1e	32.6e	1.7c
Phy 375 Trt <sup>y</sup>	14.0b	16.02a	1.1b	20.0abc	34.4a	1.7a	21.2c	39.3c	1.9b
Stv 5288 UT	14.0b	13.7d	0.98cd	19.8bc	28.8e	1.45d	20.4d	34.3d	1.7c
Stv 5288 Trt	13.0c	14.2c	1.1b	20.6ab	31.6c	1.5c	21.5bc	39.6c	1.8b
FM 1740 UT	15.0a	13.4d	0.89e	19.2c	30.3d	1.5c	20.0d	34.4d	1.7c
FM 1740 Trt	14.8a	14.1c	0.95d	19.8bc	32.1c	1.7b	21.7bc	36.1d	1.7c
Phy 499 UT	13.2c	13.2d	1.0c	20.8ab	31.8c	1.5c	18.7e	34.5d	1.8b
Phy 499 Trt	13.8b	13.5d	0.98cd	21.0a	34.5a	1.7b	22.0bc	39.5c	1.8b
Stv 5458 UT	15.0a	15.2b	1.0c	19.0c	31.8c	1.67b	22.3ab	41.5b	1.9b
Stv 5458 Trt	15.0a	15.8a	1.10b	20.0abc	33.0b	1.65b	23.1a	47.1a	2.0a
LSD(0.05)	0.3	0.5	0.04	0.7	0.6	0.04	0.6	0.8	0.04

<sup>y</sup> Trt= Aeris<sup>®</sup> seed treatment by Bayer Crop Science; UT=Untreated seed

<sup>x</sup> Average six consecutive plants destructively sampled .

<sup>w</sup> DAE=Days After Emergence.

Table 4.7	Gossypium hirsutum variety growth regarding growth parameters, total
	nodes, plant height, height to node ratio during square, bloom and open boll,
	in <i>R. reniformis</i> infested soils at the Leeper silty clay loam location.

Treatment		Square			Bloom			Open Boll		
		$(40 \text{ DAE}^{x})$	)		(70 DAE <sup>x</sup> )		(1	$(100 \text{ DAE}^{\mathrm{x}})$		
		Plant Htw	HNR		Plant Ht	HNR		Plant Ht	HNR	
	$TN^w$	(inches)	(inches)	TN	(inches)	(inches)	TN	(inches)	(inches)	
Phy 375 UT <sup>y</sup>	13.3a <sup>z</sup>	14.1de	1.0cd	17.9bc	30.6e	1.70cd	24.8a	40.7c	1.60de	
Phy 375 Trt <sup>y</sup>	13.3a	14.8b	1.10b	18.8a	33.0c	1.76c	23.5b	43.7a	1.90a	
Stv 5288 UT	13.2a	13.4fg	1.00de	17.8c	27.5g	1.54f	22.5cd	38.5d	1.70cd	
Stv 5288 Trt	12.9a	14.6bc	1.10b	18.4abc	32.2d	1.70c	22.7cd	42.5b	1.90a	
FM 1740 UT	13.2a	13.2g	0.99e	18.2abc	26.5h	1.50g	22.1d	35.7e	1.50e	
FM 1740 Trt	13.3a	13.5f	1.09de	18.8a	28.7f	1.53f	23.10bc	42.8ab	1.70cd	
Phy 499 UT	13.3a	16.4a	1.20a	18.5ab	34.1b	1.80b	24.6a	44.0a	1.80b	
Phy 499 Trt	13.5a	16.4a	1.20a	18.3abc	34.8a	1.90a	23.0bc	44.1a	1.90a	
Stv 5458 UT	13.6a	13.9e	1.00de	18.5ab	30.5e	1.65e	23.5b	40.6c	1.60de	
Stv 5458 Trt	13.3a	14.3cd	1.10c	18.8a	31.7d	1.69d	25.0a	42.1b	1.70cd	
LSD(0.050)	0.4	0.3	0.04	0.5	0.8	0.04	0.6	0.8	0.04	

<sup>z</sup> Means within columns followed by the same letter are not different according to the Least Significant Difference means separation test  $P\alpha$ =0.05.

<sup>y</sup> Trt= Aeris<sup>®</sup> seed treatment by Bayer Crop Science; UT=Untreated seed.

<sup>x</sup> DAE=Days After Emergence.

<sup>w</sup> Six consecutive plants destructively sampled.

Table 4.8Average plant height (inches) at each node culminating in total height<br/>(inches) of G. hirsutum measured during square (40 days after emergence)<br/>in R. reniformis infested soils at the Marrietta fine sandy loam location at<br/>Mississippi State University.

Treatment	Plant height at each node (inches) <sup>w</sup>											
	Node Number											
	1 <sup>x</sup>	3	5	7	9	11	13	15				
Phy 375 UT <sup>y</sup>	1.5b <sup>z</sup>	4.5ab	6.5a	9.1a	12.5a	14.4b	14.7c	14.9c				
Phy 375 Trt <sup>y</sup>	1.7b	4.4abc	6.5a	9.2a	12.5a	15.1a	15.9a	16.0a				
Stv 5288 UT	2.1a	4.5ab	6.1ab	8.3c	10.8d	12.5e	13.5ef	13.7d				
Stv 5288 Trt	2.2a	4.6a	6.2ab	8.5c	11.5bc	13.5d	14.2d	14.2c				
FM 1740 UT	1.6b	3.8d	5.5c	7.6d	10.5de	12.4e	13.2f	13.6f				
FM 1740 Trt	1.7b	3.7d	6.0b	8.4c	11.3bc	13.3d	13.8de	14.1c				
Phy 499 UT	1.8b	4.1c	5.6c	7.5ab	10.4e	12.0f	13.1f	13.2f				
Phy 499 Trt	1.7b	4.2bc	6.5a	9.0ab	11.4bc	12.7e	13.4ef	13.5d				
Stv 5458 UT	1.7b	4.4abc	6.5a	8.5c	11.2c	14.0c	14.9c	15.2b				
Stv 5458 Trt	1.8b	4.2bc	6.3ab	8.8bc	11.7b	14.4b	15.4b	15.8a				
LSD(0.05)	0.2	0.3	0.3	0.4	0.3	0.3	0.5	0.5				

<sup>y</sup> Trt= Aeris<sup>®</sup> seed treatment by Bayer Crop Science; UT=Untreated seed.

<sup>x</sup> Odd node measurements are shown to facilitate reporting.

Table 4.9Average plant height (inches) at each node culminating in total height<br/>(inches) of G. hirsutum measured during square (40 days after emergence)<br/>in R. reniformis infested soils at the Leeper silty clay loam location at<br/>Mississippi State University.

	Plant height at each node (inches) <sup>w</sup>									
Treatment	Node Number									
	1 <sup>x</sup>	3	5	7	9	11	13	15		
Phy 375 UT <sup>y</sup>	1.7cd <sup>z</sup>	4.3ab	6.6b	8.7b	11.5b	13.4c	13.9b	14.1c		
Phy 375 Trt <sup>y</sup>	1.7cd	4.4ab	6.5b	8.5b	11.2b	13.8b	14.7a	15.1a		
Stv 5288 UT	2.0bc	4.4a	6.5b	8.3b	10.7c	12.3e	12.8d	12.8e		
Stv 5288 Trt	2.3a	4.4ab	6.5b	8.6b	11.5b	12.9d	13.4c	13.4d		
FM 1740 UT	2.0b	4.1b	5.9c	7.6c	10.5d	12.5e	13.4c	13.7d		
FM 1740 Trt	2.4a	4.3ab	6.3b	8.4b	11.2b	13.3c	14.2b	14.4b		
Phy 499 UT	1.6c	3.7c	5.7c	7.8c	10.4d	11.8f	12.1e	12.1f		
Phy 499 Trt	1.8bc	3.6c	5.7c	7.7c	10.2d	12.3e	13.2c	13.4d		
Stv 5458 UT	1.9bc	4.2bc	6.2b	8.6b	11.4b	12.7de	13.3c	13.5d		
Stv 5458 Trt	2.0b	4.6a	7.2a	9.4a	12.2a	14.4a	15.0a	15.2a		
LSD(0.05)	0.3	0.3	0.3	0.4	0.3	0.4	0.3	0.3		

<sup>y</sup> Trt= Aeris<sup>®</sup> seed treatment by Bayer Crop Science; UT=Untreated seed.

<sup>x</sup> Odd node measurements are shown to facilitate reporting.

Average plant height (inches) at each node culminating in total height of G. Table 4.10 hirsutum measured during bloom (70 days after emergence) in R. reniformis infested soils at Marrietta fine sandy loam locations at Mississippi State University.

		<u>Plant</u>	height at ea	ach node (inch	es) <sup>w</sup>				
Treatment		Node Number							
	11 <sup>x</sup>	13	15	17	19	21			
Phy 375 UT <sup>y</sup>	14.5e <sup>z</sup>	19.4	24.8g	27.9e	28.8e	29.1d			
Phy 375 Trt <sup>y</sup>	20.5b	27.2a	31.5a	33.3a	34.3a	34.1a			
Stv 5288 UT	17.3d	22.6de	25.6f	28.1e	28.7e	28.7d			
Stv 5288 Trt	18.6c	22.6de	26.6e	29.9d	31.4c	31.9b			
FM 1740 UT	16.8d	22.6de	26.8e	29.9d	30.4d	30.5c			
FM 1740 Trt	21.3a	26.4b	30.4b	31.9b	32.3bc	32.3b			
Phy 499 UT	16.9d	22.2e	28.3d	30.9c	32.1bc	32.2b			
Phy 499 Trt	18.3c	23.2d	28.6d	32.4b	33.8a	34.1a			
Stv 5458 UT	18.6c	24.0c	29.5c	31.8b	32.3bc	32.3b			
Stv 5458 Trt	18.3c	24.4c	29.3c	32.1b	32.8b	33.0b			
LSD(0.05)	0.5	0.7	0.5	0.7	0.9	0.9			

<sup>z</sup> Means within columns followed by the same letter are not different according to the Least Significant Difference means separation test  $P\alpha$ =0.05.

<sup>y</sup> Trt= Aeris<sup>®</sup> seed treatment by Bayer Crop Science; UT=Untreated seed.
 <sup>x</sup> Odd node measurements <sup>used</sup> to facilitate reporting.

Table 4.11Average plant height (inches) at each node culminating in total height<br/>(inches) of G. hirsutum measured during bloom (70 days of emergence) in<br/>R. reniformis infested soils at the Leeper silty clay loam location at<br/>Mississippi State University.

		eight at each	node (inch	es) w						
Treatment		Node Number								
	11 <sup>x</sup>	13	15	17	19	21				
Phy 375 UT <sup>y</sup>	14.4f <sup>z</sup>	19.1h	24.6g	26.4g	28.8d	30.2bc				
Phy 375 Trt <sup>y</sup>	21.4a	28.1a	32.4a	33.5a	33.7a	33.7a				
Stv 5288 UT	14.7f	20.1g	25.2g	26.9g	27.5e	27.7e				
Stv 5288 Trt	18.5c	21.7f	25.3g	28.5c	29.6d	29.9cc				
FM 1740 UT	17.0f	21.7g	26.3e	29.1d	30.3c	31.9b				
FM 1740 Trt	18.9c	25.0c	29.60b	32.2b	33.1ab	33.3a				
Phy 499 UT	16.4e	22.2e	28.2d	30.1d	30.6c	30.7c				
Phy 499 Trt	20.5b	25.6b	28.8c	30.5d	30.9c	31.2bc				
Stv 5458 UT	16.9d	21.1f	26.0f	27.9f	29.0d	29.4d				
Stv 5458 Trt	18.5c	24.1d	29.6b	32.0b	32.7b	32.9a				
LSD(0.05)	0.5	0.4	0.3	0.6	0.7	0.8				

<sup>y</sup> Trt= Aeris<sup>®</sup> seed treatment by Bayer Crop Science; UT=Untreated seed.

<sup>x</sup> Odd node measurements are shown to facilitate reporting.

Table 4.12Percent (%) fruit retention<sup>u</sup> at sympodial positions 1, 2 and > 2 during<br/>square (40 DAE <sup>w</sup>), bloom (70 DAE <sup>w</sup>) and open boll (100 DAE <sup>w</sup>) at the<br/>Marrietta fine sandy loam location infested with *R. reniformis* at Mississippi<br/>State University.

		Square			Bloom			Open Boll		
Treatment	% Retention <sup>v</sup>		% Retention			9	6 Retentio	n		
	Pos 1 <sup>x</sup>	Pos 2	Pos > 2	Pos 1	Pos 2	Pos>2	Pos 1	Pos 2	Pos>2	
Phy 375 UT <sup>y</sup>	97.5a <sup>z</sup>	59.2c	24.1c	81.6b	52.9a	23.1d	33.9c	13.2c	10.0a	
Phy 375 Trt <sup>y</sup>	100.0a	70.7ab	30.6c	88.7a	51.9a	42.1b	41.7b	20.4a	2.7c	
Stv 5288 UT	98.9a	76.6a	52.7a	80.0bc	55.3a	52.2a	36.6c	9.2d	4.6b	
Stv 5288 Trt	100.0a	71.3ab	42.8b	81.2b	51.1a	32.7c	42.4b	13.3c	4.6b	
FM 1740 UT	99.2a	75.9a	52.9a	73.8cd	44.8c	22.9d	35.0c	15.0bc	0.4d	
FM 1740 Trt	100.0a	75.1a	53.9a	77.6bc	45.8bc	24.6d	37.1c	16.4b	1.5cd	
Phy 499 UT	95.9a	45.0d	30.2c	72.3d	45.9bc	19.9d	38.7bc	13.1c	2.5c	
Phy 499 Trt	100.0a	69.7ab	29.7c	88.5a	50.1ab	30.6c	47.3a	16.5b	2.2c	
Stv 5458 UT	95.9a	66.7b	51.3a	74.7cd	44.2c	19.5d	37.1c	8.7d	1.7cd	
Stv 5458 Trt	98.6a	74.0a	42.1b	81.3b	55.7a	51.3a	46.9a	12.5c	2.2c	
LSD(0.05)	3.2	5.6	6.5	3.7	4.3	5.5	3.7	2.3	1.3	
7 ) ( '.1 '	1	C 11	11 /1		1	1.0	· ·	1.	1	

<sup>y</sup> Trt= Aeris<sup>®</sup> seed treatment by Bayer Crop Science; UT=Untreated seed.

<sup>x</sup> Pos=Sympodial (fruiting) position.

<sup>w</sup> DAE=Days after Emergence.

<sup>v</sup> % retention was by fruiting position across the whole plant.

Table 4.13Percent (%) fruit retention<sup>u</sup> at sympodial positions 1, 2 and > 2 during<br/>square (40 DAE<sup>v</sup>), bloom (70 DAE<sup>v</sup>) and open boll (100 DAE<sup>v</sup>) at the<br/>Leeper silty clay loam location infested with *R. reniformis*.

	Square				Bloom			Open Boll		
Treatment	%	1	Retention <sup>w</sup>		% Retention <sup>w</sup>		% Retention <sup>w</sup>			
	Pos 1 <sup>x</sup>	Pos 2	Pos > 2	Pos 1	Pos 2	Pos >2	Pos 1	Pos 2	Pos>2	
Phy 375 UT <sup>y</sup>	70.3c <sup>z</sup>	41.1c	7.1e	67.0d	39.9d	12.1g	32.4f	18.1c	4.9d	
Phy 375 Trt <sup>y</sup>	81.5bc	52.9bc	9.4e	84.2b	48.8bc	15.0f	43.8b	23.2a	3.7e	
Stv 5288 UT	90.3ab	62.6ab	19.9bcd	76.8c	47.4c	20.8d	36.7e	17.6c	6.1c	
Stv 5288 Trt	100.0a	66.0ab	29.5b	78.1c	49.2bc	50.4a	44.8b	18.9c	5.5d	
FM 1740 UT	100.0a	71.3a	14.7de	76.2c	33.7f	24.5c	32.8f	14.1d	3.1f	
FM 1740 Trt	100.0a	75.7a	26.4bc	90.3a	59.9a	26.4c	47.7a	18.6c	7.8a	
Phy 499 UT	96.5a	69.2a	28.3b	85.5b	37.3e	25.9c	33.7f	14.3d	6.7b	
Phy 499 Trt	100.0a	70.7a	37.4a	91.1a	47.2c	38.2b	39.6d	20.9b	4.2e	
Stv 5458 UT	73.7c	42.0c	17.6cd	78.7c	40.8d	15.6f	36.7e	14.0d	5.4d	
Stv 5458 Trt	77.3bc	47.5c	26.1bc	85.4b	51.5b	17.7e	41.6c	18.4c	4.2e	
LSD(0.05)	12.5	13.1	7.7	3.5	3.0	2.4	1.6	1.2	0.6	

<sup>y</sup> Trt= Aeris<sup>®</sup> seed treatment by Bayer Crop Science; UT=Untreated seed.

<sup>x</sup> Pos=Sympodial (fruiting) position.

<sup>w</sup> % retention was by fruiting position across the whole plant.

<sup>v</sup> DAE=Days after Emergence.

Table 4.14Gossypium hirsutum maturity measured by nodes above white flower<br/>(NAWF), nodes above cracked boll (NACB) and percent open boll (open<br/>boll), as affected by variety treated with Aeris® seed treatment compared to<br/>no nematicide during bloom (70 DAEx) and open boll (100 DAEx) in R.<br/>reniformis infested soils at the Marrietta fine sandy loam location.

Treatment	NAWF <sup>v</sup>	NACB <sup>v</sup>	Open Boll <sup>v</sup>
	(Num)	(Num)	(%) <sup>y</sup>
Phy 375 UT <sup>w</sup>	8.8abc <sup>z</sup>	9.4b	10.4ef
Phy 375 Trt <sup>w</sup>	8.0h	7.3d	28.4a
Stv 5288 UT	8.7abcd	10.5a	9.3f
Stv 5288 Trt	8.5cdef	8.2c	17.5c
FM 1740 UT	9.1a	9.4b	13.5de
FM 1740 Trt	8.6bcde	9.3b	22.0b
Phy 499 UT	8.4defg	10.4a	14.7cd
Phy 499 Trt	8.2fg	7.2d	24.0b
Stv 5458 UT	8.9ab	10.8a	6.9f
Stv 5458 Trt	8.3efgh	8.9b	15.1cd
LSD(0.05)	0.3	0.8	3.8

<sup>z</sup> Means within columns followed by the same letter are not different according to the Least Significant Difference means separation test  $P\alpha$ =0.05.

<sup>y</sup> % open boll derived from open first position bolls/total number of first position bolls retained.

<sup>x</sup> DAE=Days after Emergence.

<sup>w</sup> Trt= Aeris<sup>®</sup> seed treatment by Bayer Crop Science; UT=Untreated seed.

Table 4.15 *Gossypium hirsutum* maturity measured by nodes above white flower (NAWF) (70 DAE<sup>v</sup>), nodes above cracked boll (NACB) (100 DAE<sup>v</sup>) and percent open boll (100 DAE<sup>v</sup>) as affected by variety treated with Aeris<sup>®</sup> seed treatment compared to no nematicide during bloom and open boll in *R*. *reniformis* infested soils at the Leeper silty clay loam location.

Treatment	NAWF <sup>x</sup>	NACB <sup>v</sup>	Open Boll <sup>v</sup>
	(Num)	(Num)	(%) <sup>y</sup>
Phy 375 UT <sup>w</sup>	8.2bc <sup>z</sup>	10.5a	12.3d
Phy 375 Trt <sup>w</sup>	8.0bc	7.3d	22.7a
Stv 5288 UT	8.6ab	8.3c	7.4f
Stv 5288 Trt	8.2bc	7.5d	14.6c
FM 1740 UT	8.4abc	8.9b	8.1e
FM 1740 Trt	7.9c	8.3c	18.8b
Phy 499 UT	8.9a	8.5b	12.6d
Phy 499 Trt	8.2bc	7.5d	15.3c
Stv 5458 UT	8.0bc	8.3c	6.6g
Stv 5458 Trt	7.90c	7.8d	14.7c
LSD(0.05)	0.6	0.4	0.8

<sup>*z*</sup> Means within columns followed by the same letter are not different according to the Least Significant Difference means separation test  $P\alpha$ =0.05.

<sup>y</sup> % open boll derived from open first position bolls/total number of first position bolls retained.

<sup>x</sup> Average of six consecutive plants destructively harvested.

<sup>w</sup> Trt= Aeris<sup>®</sup> seed treatment by Bayer Crop Science; UT=Untreated seed.

<sup>v</sup> DAE=Days after Emergence.

Table 4.16 Monopodial (vegetative) branch and boll production at Mississippi State University collected during open boll (100 days after emergence) comparing Aeris<sup>®</sup> seed treatment and no treatment in *R. reniformis* infested soils at the Marrietta fine sandy loam and the Leeper silty clay loam locations.

	Marrietta fine	e sandy loam	Leeper silty clay loam		
Treatment	Monopodial Branchy	Monopodial Boll <sup>y</sup>	Monopodial Branchy	Monopodial Bolly	
	(Num/plant)	(Num/plant)	(Num/plant)	(Num/plant)	
Phy 375 UT <sup>x</sup>	1.0de <sup>z</sup>	2.0d	4.0b	3.5cd	
Phy 375 Trt <sup>x</sup>	3.0ab	4.0a	5.0a	8.4a	
Stv 5288 UT	1.0e	2.0cd	3.0d	2.5d	
Stv 5288 Trt	3.0bc	2.0bcd	5.0a	3.2cd	
FM 1740 UT	2.0cde	2.0cd	3.0d	3.3cd	
FM 1740 Trt	2.0bcd	3.0bc	3.0d	3.5cd	
Phy 499 UT	2.0cde	2.0cd	4.0b	3.4cd	
Phy 499 Trt	3.0a	3.0ab	5.0a	5.7b	
Stv 5458 UT	2.0bcd	2.0bcd	3.0cd	4.4c	
Stv 5458 Trt	3.0a	3.0ab	3.0bc	6.5b	
LSD(0.05)	0.7	0.8	0.5	0.9	

<sup>y</sup> Six plants with normal terminal sampled per plot.

<sup>x</sup> UT=Untreated; TRT=Aeris<sup>®</sup> seed treatment.

Table 4.17Basal stalk and boll diameter (mm) taken at ninth and twelfth node below<br/>terminal (70 days after emergence) comparing Aeris® seed treatment<br/>compared to no nematicide in *R. reniformis* infested at the Marrietta fine<br/>sandy loam and the Leeper silty clay loam locations.

Treatment	Marrietta fine sandy	Leeper silty clay loam	5 5		dy loam	Leeper silty clay loam		
	loam							
	Basal Stalk	Basal Stalk	Boll	Boll	Boll Dia	Boll	Boll	Boll Dia
	Dia <sup>y</sup>	Dia	Dia	Dia	Diff	Dia	Dia	Diff
			Node 9 <sup>x</sup>	Node 12		Node 9 <sup>x</sup>	Node12 <sup>x</sup>	
Phy 375 UT <sup>w</sup>	9.5e <sup>z</sup>	9.8cd	17.1g	31.2bc	14.2a	27.3de	32.5de	6.7c
Phy 375 Trt <sup>w</sup>	10.7bc	10.3bcd	28.6a	32.8a	3.6f	28.0de	34.0bc	4.5d
Stv 5288 UT	9.8de	9.6d	21.3e	28.7d	7.3b	21.2g	31.7e	10.5a
Stv 5288 Trt	10.7bc	11.1ab	21.3e	31.6abc	10.3b	29.8c	33.2cd	4.3d
FM 1740 UT	10.4c	11.1ab	17.8g	32.3ab	15.0a	28.4d	34.2bc	6.4c
FM 1740 Trt	10.6bc	11.3ab	23.4d	32.3ab	8.9c	30.8bc	34.8b	3.5e
Phy 499 UT	11.2b	10.7bc	20.4e	30.8c	10.8b	31.3b	34.1bc	2.8f
Phy 499 Trt	11.3b	12.1a	26.4b	31.5abc	5.2e	32.9a	36.6a	3.6e
Stv 5458 UT	10.2c	10.4bcd	19.4f	30.7c	11.4b	23.4f	32.5de	9.1b
Stv 5458 Trt	12.3a	12.1a	24.4c	31.3bc	6.3d	26.6e	34.1bc	6.5c
LSD(0.05)	0.6	1.0	1.0	1.0	1.3	1.1	1.1	0.8

<sup>z</sup> Means within columns followed by the same letter are not different according to the Least Significant Difference means separation test  $P\alpha$ =0.05

<sup>y</sup> Diameters taken with digital calipers at boll center and cotyledonary node for stalk.

<sup>x</sup> Bolls at node 12 from terminal are the oldest boll and bolls at node 9 are the youngest.

Table 4.18Yield (Lbs Lint Cotton/Ac) of G. hirsutum varieties grown in R. reniformisinfested soils of the Marrietta fine sandy loam and Leeper silty clay loamlocations treated with Aeris<sup>®</sup> seed treatment or untreated.

Treatment	Marrietta fi	ne sandy loam	Leeper sil	ty clay loam
	Lbs Lint/Ac <sup>y</sup>	Yield Difference w	Lbs Lint/Ac <sup>y</sup>	Yield Difference w
Phy 375 UT <sup>x</sup>	1384.0f <sup>z</sup>	430.0	1482.0f	173.0
Phy 375 Trt <sup>x</sup>	1814.0a		1538.0de	
Stv 5288 UT	1482.0de	203.0	1524.0ef	94.0
Stv 5288 Trt	1685.0b		1624.0c	
FM 1740 UT	1508.0d	28.0	1580.0cd	121.0
FM 1740 Trt	1536.0cd		1624.0c	
Phy 499 UT	1457.0de	177.0	1719.0ab	167.0
Phy 499 Trt	1634.0bc		1768.0a	
Stv 5458 UT	1435.0e	60.0	1624.0c	90.0
Stv 5458 Trt	1495.0de		1689.0b	
LSD(0.05)	62.6		55.2	

<sup>y</sup> Lbs lint cotton formulated using harvested seed cotton weights x established lint % for cited varieties taken from MSU Official Variety Trials (OVT).

<sup>x</sup> Trt= Aeris<sup>®</sup> seed treatment by Bayer Crop Science; UT=Untreated seed.

<sup>w</sup> Yield differences represents difference between statistically derived varieties treated with Aeris<sup>®</sup> seed treatment nematicide and untreated check.

	Inoculated	Root Biomass (grams) <sup>y</sup>		Shoot Bi (gran	
Treatment	Population <sup>uw</sup>	Aeris <sup>®</sup> Treated	Untreated	Aeris <sup>®</sup> Treated	Untreated
FM 1740	0	55.9bc <sup>z</sup>	44.1fg	77.1ab	45.6i-l
PHY 375	0	64.5a	45.7ef	60.8d-g	53.9g-j
PHY 499	0	64.4a	44.8f	84.7a	53.0g-k
STV 5288	0	53.8bc	35.3ijk	86.3a	64.6c-g
STV 5458	0	56.8b	42.6fgh	70.9b-e	54.3g-j
FM 1740	2,500	51.4bcd	35.1ijk	63.1c-g	45.8i-1
PHY 375	2,500	50.4cde	24.91	52.4g-k	40.6klm
PHY 499	2,500	56.7b	42.3fgh	72.6bcd	59.1e-h
STV 5288	2,500	53.9bc	24.51	73.6bc	47.2h-l
STV 5458	2,500	54.1bc	39.4ghi	71.4bcd	52.7g-k
FM 1740	5,000	38.5hij	26.51	55.7f-j	35.1lm
PHY 375	5,000	25.11	12.9m	51.6g-k	42.7i-m
PHY 499	5,000	55.0bc	45.8ef	70.0b-e	51.9g-k
STV 5288	5,000	37.6ijk	25.11	54.5g-j	37.9lm
STV 5458	5,000	46.4def	27.51	67.7b-f	44.2i-l
FM 1740	7,500	35.8ijk	13.1m	53.4g-j	38.7lm
PHY 375	7,500	14.1m	8.4n	32.6m	23.2n
PHY 499	7,500	43.8fgh	13.9m	66.6b-f	42.6j-m
STV 5288	7,500	25.01	14.8m	45.3i-l	23.5n
STV 5458	7,500	25.91	13.4m	46.3i-l	25.8n
LSD(0.05)		4.0	V	7.7	7

Table 4.19Shoot and root biomass development of five G. hirsutum varieties grown in<br/>R. reniformis infested soils relative to at varying populations grown under a<br/>greenhouse environment at 90 days after emergence.

<sup>z</sup> Means within columns and rows followed by the same letter for each measured parameter are not different according to the Least Significant Difference means separation test  $P\alpha$ =0.05.

<sup>y</sup> Shoot and root biomass was acquired from the one plant grown in a 3.0 inch pot.

<sup>x</sup> Two seed per pot planted 0.5 inches deep and one removed after emergence.

<sup>w</sup> 3.0 inch pot represented 500 cc of soil.

<sup>v</sup> LSD values of 4.0 and 7.7 represent all LSDs for both columns since run together as RCB.

<sup>u</sup> *R. reniformis* added to soil at planting using a pipette via a graduated factor.

Treatment	Inoculated	Juvenile	Numbers <sup>y</sup>	Egg Nu	mbers <sup>y</sup>
	Population <sup>v,x</sup>	Aeris® Treated	Untreated	Aeris® Treated	Untreated
FM 1740	0	0.0n <sup>z</sup>	0.0n	0q	0q
PHY 375	0	0.0n	0.0n	0q	0q
PHY 499	0	0.0n	0.0n	0q	0q
STV 5288	0	0.0n	0.0n	0q	0q
STV 5458	0	0.0n	0.0n	0q	0q
FM 1740	2,500	15,991.0d	12,741.0efg	9,116.0g	5,312.0k
PHY 375	2,500	13,861.0e	11,119.0ghi	7,977.0h	2,284.0mno
PHY 499	2,500	36,729.0a	17,484.0cd	21,522.0b	15,090.0d
STV 5288	2,500	18,406.0c	16,995.0cd	13,751.0e	9,924.0g
STV 5458	2,500	18,728.0c	17,304.0cd	16,841.0c	14,678.0d
FM 1740	5,000	10,928.0ghi	6,953.0k	6,587.0ijk	6,257.0ijk
PHY 375	5,000	6,046.0kl	3,651.0m	3,600.01	1,437.0n-q
PHY 499	5,000	14,124.0e	11,866.0fgh	13,184.0e	6,850.0h-k
STV 5288	5,000	9,806.0ij	6,033.0kl	7,192.0hi	5,614.0jk
STV 5458	5,000	13,596.0ef	9,167.0j	11,621.0f	6,201.0ijk
FM 1740	7,500	8,807.0j	4,069.0m	5,377.0k	1,411.0n-q
PHY 375	7,500	3,515.0m	77.0n	876.0opq	258.0q
PHY 499	7,500	8,652.0j	4,450.0lm	5,871.0ijk	2,016.0nop
STV 5288	7,500	4,759.0lm	1,494.0n	2,446.0mn	646.0pq
STV 5458	7,500	5,253.0klm	1,622.0n	3,909.01	1,862.0nop
LSD (0.05)		1,4′	78.6 <sup>w</sup>	1,09	9.5

Table 4.20Reproduction (recovered egg and juvenile numbers) of R. reniformis across<br/>five G. hirsutum varieties treated and not treated with Aeris<sup>®</sup> seed treatment<br/>under greenhouse environments at 90 days after emergence.

<sup>y</sup> Acquired from the one plant grown in a 3.0 inch pot. Two seed per pot planted 0.5 inches deep and one removed after emergence.

x 3.0 inch pot represented 500 cc of soil.

<sup>w</sup> LSD values of 4.0 and 7.7 represent all LSDs for both columns since run together as RCB.

<sup>v</sup> *R. reniformis* added to soil at planting using a pipette via a graduated factor.

Table 4.21Plant height, total nodes and height to node ratio of five G. hirsutum<br/>varieties treated and not treated with Aeris® at varying R. reniformis<br/>populations grown under a greenhouse environment at 90 days after<br/>emergence.

Treatment	Inoculated Population <sup>vx</sup>	Plant Height (inches) <sup>y</sup>					NR ches)
	-	Aeris®	Untreated	Aeris®	Untreated	Aeris®	Untreated
FM 1740	0	24.7d-h <sup>z</sup>	21.6j-o	13.0cde	11.8f-j	2.1b-g	2.0b-j
PHY 375	0	26.1b-e	23.6f-k	13.3bcd	11.8f-j	2.2b-e	2.0b-j
PHY 499	0	28.8a	23.9e-j	13.0cde	12.0e-i	2.2bc	2.1b-f
STV 5288	0	28.0ab	23.1g-l	14.3a	12.5d-g	2.4a	2.0b-i
STV 5458	0	28.7a	23.8e-j	14.0ab	12.3e-h	2.1b-f	2.1b-g
FM 1740	2,500	23.2g-l	20.2m-r	12.3e-h	11.0ijk	2.0b-j	1.8j-p
PHY 375	2,500	23.1g-l	18.90-r	13.3bcd	11.8f-j	1.7k-p	1.7k-p
PHY 499	2,500	27.1abc	22.5h-m	13.0cde	12.0e-i	2.1b-f	2.0c-k
STV 5288	2,500	26.4bcd	22.5h-m	14.0ab	12.3e-h	2.1b-h	1.9f-m
STV 5458	2,500	26.2bcd	21.2k-p	13.8abc	11.8f-j	2.0b-j	1.9e-k
FM 1740	5,000	23.2g-l	19.8n-r	11.3hij	11.0ijk	1.9e-l	1.8h-n
PHY 375	5,000	20.91-q	16.7s	12.0e-i	11.0ijk	1.7m-p	1.4q
PHY 499	5,000	26.9a-d	22.0j-n	13.0cde	11.3hij	2.1b-g	1.9f-m
STV 5288	5,000	25.8c-f	19.2pqr	12.5d-g	11.0ijk	2.0b-j	1.6opq
STV 5458	5,000	26.8a-d	21.3j-o	13.0cde	11.8f-j	2.0b-j	1.9f-m
FM 1740	7,500	21.6j-o	19.4o-r	11.3hij	10.8jk	1.8i-o	1.7k-p
PHY 375	7,500	19.8n-r	14.3s	12.0e-i	10.01	1.6n-q	1.4q
PHY 499	7,500	25.4c-g	21.6j-o	13.0cde	11.3hij	2.0c-k	1.8h-n
STV 5288	7,500	23.9e-j	18.3r	11.0ijk	11.0ijk	1.7l-p	1.5pq
STV 5458	7,500	24.7d-l	20.91-q	12.3e-h	10.3kl	1.9e-l	1.8g-n
LSD (0.05)		1.	.5 <sup>w</sup>		0.6		0.2

<sup>y</sup> Plant height and total nodes was acquired from the one plant grown in a 3.0 inch pot. Two seed per pot planted 0.5 inches deep and one removed after emergence.

x 3.0 inch pot represented 500 cc of soil.

<sup>w</sup> LSD values of 4.0 and 7.7 represent all LSDs for both columns since run together as RCB.

<sup>v</sup> *R. reniformis* added to soil at planting using a pipette via a graduated factor.

Treatment	Inoculated Population <sup>vx</sup>	NFFB <sup>y</sup>		Basal Stalk Diameter (mm) <sup>v</sup>	
		Aeris® Treated	Untreated	Aeris <sup>®</sup> Treated	Untreated
FM 1740	0	7.0i-l <sup>z</sup>	8.3fgh	6.5e-i	5.6k-n
PHY 375	0	6.3kl	7.8hij	6.7c-g	5.1no
PHY 499	0	8.0ghi	9.0d-g	7.1bcd	6.5e-i
STV 5288	0	6.01	7.0i-1	7.7a	6.0h-i
STV 5458	0	6.8jkl	8.8e-h	7.5ab	6.4e-i
FM 1740	2,500	8.0ghi	9.3def	6.4e-i	5.6k-n
PHY 375	2,500	6.8jkl	8.33fgh	6.6d-i	4.7op
PHY 499	2,500	8.0ghi	9.0d-g	7.0b-e	5.9i-1
STV 5288	2,500	6.01	7.0i-1	7.2bc	5.5mn
STV 5458	2,500	7.0i-l	9.0d-g	6.5e-i	6.2f-j
FM 1740	5,000	8.0ghi	9.5cde	6.4e-i	5.4lmn
PHY 375	5,000	7.0i-1	10.0bcd	6.6d-i	4.6op
PHY 499	5,000	8.3fgh	10.3bc	6.4e-i	5.6k-n
STV 5288	5,000	6.5kl	9.8cde	6.3f-j	5.3mn
STV 5458	5,000	7.3ijk	8.0ghi	6.4e-i	6.1g-k
FM 1740	7,500	9.8cde	10.0bcd	6.0h-l	4.7op
PHY 375	7,500	7.0i-1	11.3a	6.4e-i	4.3p
PHY 499	7,500	8.8e-h	10.0bcd	6.4e-i	5.3mn
STV 5288	7,500	6.8jkl	10.0bcd	6.5e-i	4.4p
STV 5458	7,500	7.8ĥij	9.3def	6.5e-i	5.4Îmn
LSD(0.05)		0.6 <sup>w</sup>		0.4	

Table 4.22Node of first fruiting branch and basal stalk diameter of five G. hirsutum<br/>varieties treated and not treated with Aeris® at varying populations grown<br/>under a greenhouse environment at 90 days after emergence.

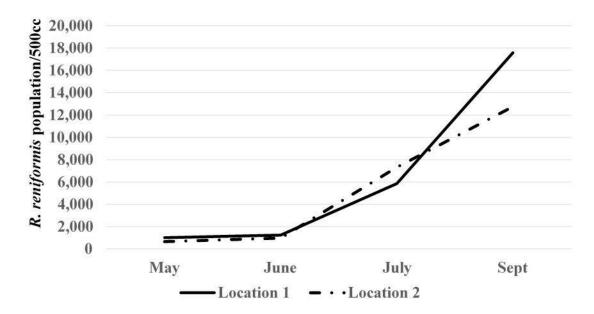
<sup>z</sup> Means followed by same letter are not different according to Least Significant Difference means separation test  $P\alpha$ =0.05.

<sup>y</sup> Node of first fruiting branch and basal diameter were acquired from the one plant grown in a 3.0 inch pot. Two seed per pot planted 0.5 inches deep and one removed after emergence.

x 3.0 inch pot represented 500 cc of soil.

<sup>w</sup> LSD values of 4.0 and 7.7 represent all LSDs for both columns since run together as RCB.

<sup>v</sup> *R. reniformis* added to soil at planting using a pipette via a graduated factor.



- Figure 4.1 Seasonal progression of *R. reniformis* populations sampled during May (atplanting), June (square), July (bloom) and August (open boll) across five *G. hirsutum* varieties (Stv 5458 B2RF, Stv 5288 B2RF, FM 1740 B2RF) grown at Mississippi State University locations across varieties.
- <sup>2</sup> Samples acquired on a per plot basis and averaged across all plots to display population dynamics of *R. reniformis* at each cotton growth stage.
- <sup>y</sup> Six samples per plot were acquired using a fluted probe from six inches from the row middle in a manner to obtain three samples from each of the two rows plots.
- <sup>x</sup> Sample depth was approximately three inches deep per sample.
- <sup>w</sup> Samples were bagged and cooled away from direct sunlight until sample processing using the elutriator/centrifuge system.

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## CHAPTER V

## CONCLUSIONS

Reniform nematode (*Rotylenchulus reniformis* Linford and Oliveira) infests 36% of the Mississippi cotton acres causing a \$130 million national loss annually. *Rotylenchulus reniformis* was previously controlled using at-planting treatments of Temik 15G or other soil fumigants. With Temik 15G being removed from the market and fumigant expense increasing, there was need to evaluate integrated crop management options involving Nematicide Seed Treatment (NST) efficacy with and without foliar applications of Vydate C-LV<sup>®</sup> and the role of commercially available varieties treated with Aeris<sup>®</sup> relative to tolerance in *R. reniformis* infested soils.

In greenhouse and field studies at the R. R. Foil Plant Science Research Center of Mississippi State University in Starkville, Mississippi, and the Tennessee Valley Research and Extension Center (TVREC) of Auburn University (AU) in Belle Mina, Alabama, effects of *R. reniformis* populations upon growth and development of Phy 375 were assessed. Of the NSTs tested in the greenhouse, Aeris<sup>®</sup> + Votivo<sup>®</sup> provided better nematode management for plants in terms of shoot and root biomass compared to Aeris<sup>®</sup> at higher nematode populations. Plants treated with Aeris<sup>®</sup> + Votivo<sup>®</sup> did maintain comparable shoot biomass in comparison to plants treated with Temik 15G, but root mass was reduced, suggesting nematode populations were impacting those roots. Field plant mapping at MSU indicated node of first fruiting branch (NFFB) was reduced and plant 169

height (PH) and height to node ratio (HNR) at open boll was increased for all plants treated with nematicides. Vydate C-LV<sup>®</sup> applications improved performance of nematicide treatments on plants at open boll in regards to PH, but plant HNR exhibited no clear advantage. Field plant mapping at TVREC also indicated that NFFB of plants was reduced and PH and HNR of plants at open boll increased with all nematicides, but PH and HNR of plants indicated no clear advantage due to Vydate C-LV<sup>®</sup> applications. At the final MSU evaluation, Vydate C-LV<sup>®</sup> improved retention of key fruiting sites (Pos 1 and Pos 2 measured 100 DAE), improved harvest maturity (Pos 1 and Pos 2 measured 100 DAE) and improved yields (Lb Lint/Ac) of plants treated with Aeris<sup>®</sup> or Aeris<sup>®</sup> + Votivo<sup>®</sup>NSTs making them equal or superior to plants grown using Temik 15G. These results suggest that producers do have viable NST options with the loss of Temik 15G.

Commercial variety tolerance to *R. reniformis* is important since no true resistance exists today in the industry. Little tolerance to *R. reniformis* has been reported in *G. hirsutum* varieties, however, studies indicate some varieties perform better than others in *R. reniformis* infested soils. Greenhouse studies indicated that plants treated with Aeris<sup>®</sup> almost always had increased root and shoot biomass, PH, TN, and basal stalk diameters with reduced egg and juvenile nematode counts and NFFB numbers compared to untreated plants, regardless of variety. However, there were many varietal differences in growth in response to nematode populations. Untreated varieties had lower fruit retention delaying harvest maturity, displayed as greater number of nodes above cracked boll, lower percent open boll and greater boll diameter differences. Some commercial varieties (Phy 499, Stv 5458 and FM 1740) evaluated showed good tolerance in *R. reniformis* infested soils.

At low to moderate *R. reniformis* populations, it is possible to use tolerant *G. hirsutum* varieties without a nematicide and reduce production costs. However, this data does agree with other findings where nematicides can be beneficial despite some varietal tolerances and can have a synergistic effect. Additionally, NST's have performed very well when compared to Temik 15G. A producer needs to understand how varietal characteristics and NST's affect yield, know their nematode species and population, and use sound agronomic practices to have an integrated and successful nematode management plan.