

MANAGEMENT OF SUGARCANE APHID, *Melanaphis sacchari* (HEMIPTERA:  
APHIDIDAE), IN SUSCEPTIBLE AND RESISTANT GRAIN SORGHUM

A Dissertation

by

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Submitted to the Office of Graduate and Professional Studies of  
Texas A&M University  
in partial fulfillment of the requirements for the degree of

DOCTOR OF PHILOSOPHY

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May 2021

Major Subject: Entomology

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## ABSTRACT

Sugarcane aphid, *Melanaphis sacchari* (Hemiptera: Aphididae), was first detected on grain sorghum, *Sorghum bicolor*, in the United States in 2013. The spread of sugarcane aphid across the sorghum-producing regions of North America necessitated increased understanding of damage by and methods for mitigation of the pest. In response, field experiments were conducted to develop economic thresholds for sugarcane aphid. Grain sorghum yield—aphid population density relationships were used to calculate economic injury levels and economic thresholds. Economic injury levels ranged from 37 and 102 aphids per leaf, and an economic threshold of 40 aphid per leaf was deemed prudent to use across the observed range of hybrid, environmental, and market conditions. Subsequently, a tally-based threshold was considered by evaluating the infestation proportion – aphid density relationship for tallies of >25, >50, and >100 aphids per leaf. Regressions showed a second order polynomial relationship yielded decisions most similar to use of the density-based threshold. The tally threshold required half the time to sample 100 leaves compared to the density-based approach. With increased introduction of grain sorghum hybrids partially resistant to sugarcane aphid, field evaluations of the grain yield–aphid population relationship were conducted across growing seasons, locations, and hybrids believed to vary in aphid susceptibility. These data verified previously established economic injury levels ranging from 27 to 72 aphids per leaf for the most susceptible hybrids. For 47 of the 49 partially resistant hybrid location-years, yield loss attributable to aphid density was not detected under

aphid densities up to 352 aphids per leaf. Population doubling time for sugarcane aphid on partially resistant grain sorghum hybrids was approximately two times that of susceptible hybrids. Finally, spray tips were evaluated for canopy penetration and coverage using grower spray equipment and two spray volumes. There were no differences in coverage among spray tips or between spray volumes. When guided by economic thresholds for susceptible hybrids, several configurations of spray equipment and volumes are effective. Overall, sugarcane aphid can be managed with use of partially aphid-resistant hybrids and aphid-susceptible hybrids with addition of insecticides applied with common grower equipment and guided by use of economic thresholds.

## DEDICATION

I dedicate my dissertation to my wife, Jennifer, for her continued support and believing in me, even when I did not. I am forever grateful for her love and support in this endeavor and in all other things we do. I also dedicate this to my children, Mason, Drew, and Tyler, for their patience throughout this process and their moral support. I love you all.

## ACKNOWLEDGMENTS

I thank my advisors and committee members: Michael Brewer, M.O. Way, Monty Dozier, and Craig Coates for their support throughout this process. Dr. Brewer has been a great supporter and reliable resource as he has stewarded me through this process and I am extremely appreciative of his patience and attention to detail. Dr. Way has been a great motivator and champion of my successes and I am grateful for his support. Dr. Dozier has supported my growth since before this endeavor and has graciously continued to do so through many changes, all along with a strong and kind spirit. Dr. Coates stepped up to fill in late and has provided thought provoking questions and input to help polish some of the finer points.

I would also like to thank Dr. Kevin Heinz for his early input regarding my plan of research, and the Department of Entomology faculty and staff for their flexibility in having me as a Ph.D. student working remotely. Special thanks to collaborators who participated in the experimentation: David Buntin, Francis Reay-Jones, Nick Seiter, David Kerns, and Robert Bowling. Special thanks also to the individuals who helped in plot maintenance and data collection at the various trial locations – this would not have been possible without their assistance. Special thanks also to the members of the Fort Bend Row Crops Committee: Paul and Linda Freund, Alan and Lisa Stasney, Kenny Poehls, and Mark Wleczyk for their support during my time as Fort Bend County Extension Agent. I also thank my AgriLife Extension supervisors for allowing me to begin this process, and my Syngenta managers for supporting the completion of my dissertation. I also wish to thank industry partners for materials and funding support.

## CONTRIBUTORS AND FUNDING SOURCES

### **Contributors**

This work was supervised by a dissertation committee consisting of Dr. Michael Brewer (Committee Chair), Dr. M.O. Way (Committee Co-Chair), Dr. Craig Coates (Department of Entomology), and Dr. Monty Dozier (Department of Soil and Crop Sciences). Portions of data used in Chapters 2 and 4 were provided by Dr. David Buntin (Georgia), Dr. Nick Seiter (Arkansas, Illinois), Dr. Francis Reay-Jones (Clemson), Dr. David Kerns (Louisiana State, Texas A&M). Data collection at Texas locations was assisted by D. Anderson, K. Fuhrmann, D. Olsovsky, T. Ahrens, L. Pruter, L. DeLeon, A. Reyes, J. Glover, I. Esquivel, A. Farris, M. Killian, J. Grunseich, Z. Shihabuddin, S. Fennessy, D. Locke, G. Scott, and C. Dickson. All other work conducted for the dissertation was completed by the student independently.

### **Funding Sources**

This work made possible in part by the Texas Grain Sorghum Board and the United Sorghum Checkoff Program. It was also supported by USDA-NIFA (Crop Protection and Pest Management Competitive Grants Program, Applied Research and Development, 2014-70006-22528). This work was also supported by the Texas Rice Research Foundation and was made possible in part by funding and material support from Industry partners including: Advanta, BH Genetics, Certis, DeKalb, Dyna-Gro, FMC, Sorghum Partners, Terral, Warner, Bayer, BASF, Corteva, Syngenta, TeeJet Technologies, and Greenleaf Technologies.

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## 1. INTRODUCTION

### 1.1. Introduction

Grain sorghum, *Sorghum bicolor* L., is an important crop in the United States. In 2015, there were 7.7 million acres harvested in the U.S., an increase of 21% from 2014 (USDA-FSA 2015). The key insect pests of sorghum include several aphid species, sorghum midge, headworms, and stinkbugs. Economic thresholds and methods for chemical and cultural control of these pests are well known (Knutson et al. 2018). Sugarcane aphid, *Melanaphis sacchari* Zehntner (Hemiptera: Aphididae), has been an economically important pest of sorghum in parts of Asia and Africa for several decades (Singh et al. 2004). It was first found in the continental United States on sugarcane in Florida in 1977 and was found on sugarcane in Louisiana in 1999 (Mead 1978, Denmark 1988, White et al. 2001). While Denmark (1988) also reported that *M. sacchari* in Florida would feed on *Sorghum* spp., it was not considered a pest until the recent outbreak on sorghum was first detected along the Texas Gulf Coast in 2013 (Villanueva et al. 2014). In 2013, this new pest of grain sorghum was detected in 38 counties and parishes in Texas, Louisiana, Oklahoma, and Mississippi (Villanueva et al. 2014). Confirmed sugarcane aphid populations increased to 12 states and more than 300 counties in 2014 and 17 states and more than 400 counties in 2015 (Bowling et al. 2016).

Previous introductions of aphid pests of cereal grains have been documented. Two such examples are Russian wheat aphid, *Diuraphis noxia* Kurdjumov, and greenbug, *Schizaphis graminum* Rondani (Peairs and Quisenberry 1998, Michels and Burd 2007). Integrated pest management (IPM) strategies for managing these pests

included adoption of thresholds for determining timing of chemical control as well as use of resistant cultivars and hybrids and recognition of aphid natural enemies (Royer et al. 2015).

One major consideration for management strategies is the population growth potential of sugarcane aphid. Based on field observations, there is considerable variation in population growth depending on host plant species and plant genetic background. Da Silva et al. (2014) reported a population doubling time of 2.3 days on sweet sorghum, while Akbar et al. (2010) reported doubling times of 4.5 and 13.9 days on susceptible and resistant sugarcane, respectively. Additionally, *M. sacchari* is known to persist on other *Sorghum* spp, including Johnsongrass, *Sorghum halapense*, a grass commonly found in pastures, right of ways, and roadsides. Other plants reported to support the aphid include grasses in the genera *Saccharum*, *Oryza*, *Panicum*, and *Pennisitum* (Singh et al. 2004).

The economic injury level is the lowest insect population density that will cause economic damage. The economic threshold is a population density below the economic injury level that should trigger a management tactic to reduce populations and prevent economic damage (Pedigo, 1999). Pedigo's formula for economic injury level is  $EIL = C/(V*I*D*K)$  where C is the control cost, V is the value of the crop, K is the proportion of the insect population controlled, I is injury units per insect per production unit, and D is damage per unit injury. The economic threshold is a point somewhere below the economic injury level that allows time for the control tactic (typically an insecticide) to be used prior to incurring plant injury that causes economic damage. Pedigo (1999) also

provides for a descriptive economic threshold where insect population growth can also be considered.

With the increasing availability and regional adaptation of resistant hybrids, adjustments to economic thresholds originating from hybrids susceptible to sugarcane aphid may need to be considered and evaluated. Teetes (1994) explained that when greenbug resistant sorghum hybrids were introduced, there was less yield loss per greenbug and more greenbugs were required for equivalent plant damage to resistant hybrids compared to susceptible hybrids. However, because the economic threshold was based on plant damage, it was not different for resistant and susceptible hybrids. In contrast, when plant susceptibility is linked to aphid population estimates, and aphid population estimates are used for decision making, a change in hybrid susceptibility may also lead to a change in the yield-aphid density relationship. Adjusting thresholds based on *M. sacchari* populations should be considered for susceptible and resistant hybrids.

## **1.2. Rationale and Significance**

The first confirmed detection of sugarcane aphid on sorghum in Texas was in 2013. Since that time, it has been found in 17 states, including all of the major sorghum-producing states (Bowling et al. 2016). *M. sacchari* has the potential to overwinter on volunteer grain sorghum, forage sorghums and Johnson grass, which persist during the winter in South Texas and Mexico (Bowling et al. 2016). This overwintering, along with wind-aided migration, gives the aphid the potential for rapid colony establishment and expansion in commercial grain sorghum fields. Preliminary data suggested significant

yield decline occurred at population levels between 50 and 125 aphids/leaf (Brewer et al. 2017). This range provided some flexibility for frequency of scouting and delay between scouting and application. However, data indicated unexplained variability among locations. Additional research was warranted to formally calculate economic thresholds based on susceptibility of hybrids, aphid population growth, and cost of control.

The information derived from the experiments outlined in the objectives below were intended to contribute to a comprehensive guide for the management of sugarcane aphid in grain sorghum in the southern United States. The major goals of my dissertation were to 1) Evaluate the aphid population-yield loss relationship and aphid population growth potential of aphid-susceptible sorghum hybrids for use in estimating economic injury level and thresholds, 2) Evaluate tally thresholds as an alternative to aphid density-based thresholds, 3) Evaluate partially aphid-resistant sorghum hybrids for aphid growth and yield stability and 4) Evaluate selected insecticides and insecticide application technologies for selective use as guided by economic thresholds.

### **1.3. Study Area**

Working cooperatively with other university researchers, experiments to evaluate aphid susceptibility of grain sorghum hybrids were performed at multiple locations in Texas and selected locations across the southern U.S., including primary research locations at the Corpus Christi Research and Extension Center and in commercial grain sorghum fields near Rosenberg, Texas. Other locations included



Gainesville, Texas; Winnsboro, Louisiana; Monticello, Arkansas; Florence, South Carolina; and Griffin, Georgia.

#### **1.4. Objective 1: Establish sugarcane aphid thresholds for aphid-susceptible sorghum hybrids**

Following first detection of sugarcane aphid in 2013, yield loss from plant damage caused by *M. sacchari* and harvest issues related to honeydew accumulation on harvest equipment were observed across wide swaths of sorghum production areas in Texas and Louisiana (Bowling et al., 2016). In 2014 and 2015, the area experiencing damaging populations of sugarcane aphid on grain sorghum expanded dramatically. In response to the detection in 2013, an initial experiment to characterize yield loss response to damage from sugarcane aphid was conducted in 2014 at Corpus Christi, Texas and Winnsboro, Louisiana (Brewer et al. 2017). These data showed yield decline at aphid levels between 50 and 250 aphids per leaf.

To further investigate those initial findings, known susceptible sorghum hybrids were planted at various locations in Texas and throughout sorghum production regions of the southern U.S. Naturally occurring aphid populations were allowed to colonize plots and populations were manipulated using insecticides to obtain a range of sugarcane aphid population densities. Yields were recorded and the aphid density (maximum populations and cumulative aphid days) – yield relationship were evaluated to evaluate hybrid susceptibility. Aphid population growth was monitored and population doubling time calculated (Akbar et al. 2010). These data were used to calculate economic injury

levels and economic thresholds (Pedigo 1999) for use in management of sugarcane aphid in susceptible grain sorghum.

### **1.5. Objective 2: Binomial-based tally thresholds as an alternative to aphid density-based thresholds in sorghum**

Following estimation of economic injury level and economic threshold for susceptible hybrids, I evaluated the use of a binomial-based tally threshold as an alternative to a density-based threshold. For this, I utilized data from Texas locations collected in Objective 1. For each sampling date and plot, the mean aphid population for the 20 sampled leaves was calculated, along with the proportion of leaves with >25, >50, and >100 aphids per leaf (tally threshold). Regression analysis was performed for each location-year with infestation proportion as the dependent variable and the mean aphid density as the independent variable, to evaluate what proportion of leaves at each tally threshold was most representative of the economic threshold. For validation, the chosen tally threshold was compared to the density-based threshold by sampling aphid-infested sorghum fields.

### **1.6. Objective 3: Field assessment of aphid doubling time and yield of grain sorghum susceptible and partially resistant to sugarcane aphid**

Pest resistant or tolerant germplasm of any given crop can be an important component of an effective integrated approach to management of a pest, and the sugarcane aphid – grain sorghum dynamic is no different. Some sorghum hybrids with

greenbug resistant traits, as well as additional sources of resistance, show reduced damage when infested with *M. Sacchari* (Armstrong et al. 2015, Mbulwe et al. 2015). In initial experiments, ATx2752 x RTx2783, which was resistant to greenbug (Peterson et al. 1984) was used, along with a susceptible hybrid. This hybrid showed reduced populations of sugarcane aphid and subsequently, reduced yield loss, when compared to a susceptible hybrid of similar lineage (Brewer et al. 2017).

Similar to objective one, purported aphid-resistant or tolerant sorghum hybrids were planted at various locations in Texas and throughout sorghum production regions of the southern U.S. Naturally occurring aphid populations were allowed to colonize plots and populations were manipulated using insecticides to obtain a range of *M. sacchari* densities. Yields were recorded and the aphid intensity – yield relationship were estimated and economic injury levels for susceptible hybrids from objective one were confirmed. Aphid doubling time of susceptible hybrids was found to be greater than that of partially resistant hybrids, and yield loss was not detected in partially resistant grain sorghum hybrids.

## **1.7. Objective 4: Insecticide efficacy and spray application considerations**

### **1.7.1. Assess available insecticides for efficacy and residual activity in management of sugarcane aphid.**

The performance of several insecticides or insecticide combinations in selected formulations and rates were evaluated for demonstration purposes. Commercial sorghum fields with a known susceptible hybrid were used to evaluate selected commercially-

available (labeled and not labeled for *Melanaphis sacchari*) insecticides. Pre-treatment counts were taken and post treatment counts were performed every three to seven days for two to three weeks. Aphid sampling was conducted as described in objective one. Insecticides providing both fast-acting and residual efficacy were observed.

### **1.7.2. Evaluate the effect of different spray tips and total spray volume on the efficacy of selected insecticides.**

Sugarcane aphid populations colonize the underside of leaves. Often, the first large colonies found in any given field are at the base of lower leaves. Although the two commonly used insecticides available for sugarcane aphid management (sulfoxaflor, Transform WG, Dow Agrosiences and flupyradifurone, Sivanto Prime, Bayer) are systemic, they only move from the base of the plant outward. As such, when control measures are initiated, it is important to cover as much of the canopy as possible. Differences in spray coverage and canopy penetration may be important points of consideration for expected efficacy of an insecticide application. Different spray tips (Airmix, TTJ60, dual fan, turbodrop dual fan, twinjet, 30/70 air induction, and hollow cone) and spray volumes (65 and 112 L/ha) were evaluated using grower equipment and water sensitive cards to assess canopy penetration using different spray tips and spray volumes. Water sensitive cards were placed at four canopy positions with different spray tips oriented over three rows. Grower equipment was used to make applications at two spray volumes. Cards were evaluated for percent coverage using a scanner and

appropriate software (DepositScan, USDA-ARS) to consider if spray tip selection or spray volume increased spray coverage at different canopy levels.

## **1.8. Conclusion**

Since 2013, sugarcane aphid has become the most important pest of grain sorghum in the southern United States. Proper management of this pest to mitigate economic loss is important to keep grain sorghum as a viable low-risk crop used for rotation with cotton, soybean, corn, and rice. Chapter 2 of my dissertation evaluates the relationship between aphid populations and yield loss, aphid population growth in the field to estimate economic injury level and economic threshold for use across sorghum-growing regions of the southern United States. Chapter 3 investigates the use of a tally-based threshold as an alternative to a density-based threshold to provide a scouting approach that will be more time efficient. Chapter 4 investigates aphid population growth and yield stability of sorghum hybrids purported to be aphid-resistant. Finally, chapter 5 examines efficacy and residual activity of insecticides as well as potential benefits of spray tip selection and spray volume to best control sugarcane aphid. Together, these studies will contribute to a comprehensive guide for management of sugarcane aphid in grain sorghum and provide insight and add to the case studies of adaptive integrated pest management to new and introduced crop pests.

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## 2. DEVELOPMENT OF ECONOMIC THRESHOLDS FOR SUGARCANE APHID, *Melanaphis sacchari*, (HEMIPTERA: APHIDIDAE) IN SUSCEPTIBLE GRAIN SORGHUM HYBRIDS\*<sup>1</sup>

### 2.1. Introduction

Grain sorghum, *Sorghum bicolor* L., is an important crop in the southern United States of America (U.S.A). From 2012 to 2017, between 2.0 and 3.2 million hectares of grain sorghum were harvested annually in the U.S.A. (USDA-NASS 2018). The key insect pests of sorghum include several aphid species, sorghum midge (*Contarinia sorghicola* Coquillett) (Diptera: Cecidomyiidae), headworms (*Helicoverpa zea* Boddie, *Spodoptera frugiperda* J.E. Smith [Lepidoptera: Noctuidae] and *Nola sorghiella* Riley [Lepidoptera: Nolidae]), and stink bugs (Hemiptera: Pentatomidae). Economic thresholds and methods for chemical and cultural control of these pests are known (Cronholm, et al. 2007, Trostle and Fromme 2010). Sugarcane aphid, *Melanaphis sacchari* Zehntner (Hemiptera: Aphididae), has been a pest of sorghum in parts of Asia and Africa for several decades (Singh et al. 2004), but economic thresholds to guide insecticide use for sugarcane aphid control has not been considered in North America prior to the 2013 outbreak on sorghum (Brewer et al. 2019).

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<sup>1</sup> Reprinted with permission, Gordy, J.W., M.J. Brewer, R.D. Bowling, G.D. Buntin, N.J. Seiter, D.L.Kerns, F.P.F. Reay-Jones, and M.O. Way. 2019. Development of economic thresholds for sugarcane aphid (Hemiptera: Aphididae) in susceptible grain sorghum hybrids. *J. Econ. Entomol.* 122: 1251–1259.

Sugarcane aphid was first found in the continental U.S.A on sugarcane in Florida in 1977 and was found on sugarcane in Louisiana in 1999 (Mead 1978, Denmark 1988, White et al. 2001). While Denmark (1988) reported that sugarcane aphid would feed on *Sorghum* spp., it was not considered a significant sorghum pest until the recent outbreak on sorghum that was first detected along the Texas Gulf Coast in 2013 (Bowling et al. 2016). In 2015, confirmed sugarcane aphid populations on sorghum extended to 17 states and more than 400 counties (Bowling, et al. 2016). Nibouche et al. (2018) reported that this population exhibited low genetic diversity and consists of a dominant clonal lineage, MLL-F, which colonizes *Sorghum* spp. and sugarcane. It was a new invasive genotype, likely introduced into the Americas from either Africa or Asia, with Asia being the most probable source.

*Melanaphis sacchari* is an anholocyclic, parthenogenic, viviparous species, with adults either winged (apterous) or wingless (alate) (Bowling et al. 2016, Singh et al. 2004). They are 1.1 to 2.0 mm in length and can vary in color from pale yellow to gray or brown, with dark cornicles, tarsi, and antennae (Bowling et al. 2016, Villanueva et al. 2014, Blackman and Eastop 1984). Sugarcane aphids have a tremendous potential for population growth and do well in tropical and subtropical environments (Akbar 2010, Singh 2004). Sugarcane aphids feed on the underside of leaves removing large amounts of plant fluids and exuding honeydew which is deposited on lower leaves. Infested plants can exhibit stress symptoms including yellowing leaves, die-back, stunting, and failure of panicle emergence. However, there is no evidence of toxin produced by sugarcane aphid. Infestations on pre-boot sorghum, through grain development, can

cause significant yield loss. Additionally, harvest related issues caused by accumulation of honeydew on harvest equipment have been reported (Bowling et al. 2016, Knutson et al. 2016, Villanueva et al. 2014). Several species of natural enemy predators and parasitoids have been observed in sugarcane aphid-infested sorghum. These include lady beetles (Coleoptera: Coccinellidae), lacewing (Neuroptera: Hemerobiidae and Neuroptera: Chrysopidae), syrphid fly (Diptera: Syrphidae), *Aphelinus sp.* (Hymenoptera: Aphelinidae), and *Lysiphlebus testaceipes* (Hymenoptera: Braconidae) (Bowling et al. 2016, Brewer et al. 2018).

Previous introductions of aphid pests of cereal grains have been documented in the U.S.A. Two such examples are Russian wheat aphid, *Diuraphis noxia* Kurdjumov, and greenbug, *Schizaphis graminum* Rondani (Quisenberry and Peairs 1998, Michels and Burd 2007). Integrated pest management (IPM) strategies for managing these pests include adoption of thresholds for determining timing of chemical control as well as aphid-resistant plant varieties. Aphid-resistant varieties are typically not available during initial phases of aphid invasion and expansion. Therefore, judicious insecticide use is especially important during this period, as guided by economic sampling procedures, thresholds, and insecticide choice to minimize natural enemy kill (Pedigo 1999).

As applied to management of sugarcane aphid on grain sorghum, the economic injury level is the lowest sugarcane aphid population level at which economic loss of grain yield is equal to control costs. The economic threshold is the population level below the economic injury level that prompts use of an insecticide or alternative management tactic to prevent the population from exceeding the economic injury level

(Pedigo 1999). The economic injury level and threshold are potentially affected by sorghum hybrid sensitivity and environmental influences that affect the yield—aphid population relationship, as well as management considerations such as cost of insecticide, crop value, and efficacy of control. The economic threshold should also consider the lag time needed to initiate a management tactic and aphid population growth potential (Pedigo 1999).

To provide guidance on insecticide use during initial phases of sugarcane aphid invasion and expansion on sorghum in North America, field evaluations of the relationship between grain yield and aphid population estimates were conducted across multiple susceptible hybrids and a range of environmental conditions. These evaluations were repeated across years and locations. These data, along with consideration of aphid population growth potential and market factors, were used to calculate economic thresholds and propose an economic threshold most applicable to a range of hybrid, environmental, and market conditions.

## **2.2. Methods and Materials**

### **2.2.1. Experimental Design and Manipulation**

A field experiment was conducted 15 times at various locations (Texas, Louisiana, Arkansas, Georgia, and South Carolina) across the southern U.S.A. in 2014, 2015, and 2016 (Table 2-1). Known aphid-susceptible hybrids were planted in plots ranging in length from 9.14 to 12.19 m by four rows, with row spacing of 0.76 to 1.02 m. A mid- to late-planting time based on local standards was used to maximize chances

**Table 2-1 Hybrid, source, planting date, target aphid densities, irrigation, and insecticide used in regional field experiments on sorghum hybrids susceptible to sugarcane aphid in the southern U.S.A., 2014-2016; Reprinted with permission from Gordy et al. (2019)**

Year	Location	Hybrid <sup>a</sup>	Source <sup>a</sup>	Planting Date	Irrigation	Target Aphid Densities <sup>b</sup>	Insecticide(s)	Insecticide Rate <sup>c</sup>	Spray Method
2014	Corpus Christi, TX	Tx2752/Tx430	TAMU	Apr 11	dryland	50, 100, 250, 500, UTC	Sulfoxaflor	70 g/ha	CO <sub>2</sub> Backpack
2014	Winnsboro, LA	Tx2752/Tx430	TAMU	Jun 3	dryland	50, 100, 250, 500, UTC	Sulfoxaflor	70 g/ha	Self-propelled plot sprayer
2015	Corpus Christi, TX	Dekalb, DKS 53-67	Monsanto	May 2	dryland	50, 125, 250, 500, UTC	Sulfoxaflor	70 g/ha	CO <sub>2</sub> Backpack
2015	Winnsboro, LA	Tx2752/Tx430	TAMU	May 29	furrow	50, 125, 250, 500, UTC	Sulfoxaflor	70 g/ha	Self-propelled plot sprayer
2015	Rosenberg, TX	Dekalb, DKS 53-67	Monsanto	Jul 16	flood, dryland	50, 125, 250, 500, UTC	Sulfoxaflor	70 g/ha	CO <sub>2</sub> Backpack
2015	Monticello, AR	Pioneer, 83P99	Pioneer	Jun 9	furrow	25, 50, 125, 250, 430, 500, UTC	Sulfoxaflor	70 g/ha	Self-propelled plot sprayer
2015	Griffin, GA	SSC SS800A	SSC	Jun 15	sprinkler	0, 50, 125, 250, 500, UTC	Sulfoxaflor	70 g/ha	CO <sub>2</sub> Backpack
2016	Corpus Christi, TX	Dekalb, DKS 53-67	Monsanto	May 3	dryland	50, 125, 300, UTC	Sulfoxaflor, Flupyradifurone	70 g/ha, 292 ml/ha	CO <sub>2</sub> Backpack
2016	Winnsboro, LA	Terral, Rev8782	Terral	May 18	furrow	50, 125, 300, UTC	Flupyradifurone	292 ml/ha	Self-propelled plot sprayer
2016	Rosenberg, TX	Dekalb, DKS 53-67	Monsanto	May 5	dryland	50, 125, 300, UTC	Sulfoxaflor, Flupyradifurone	70 g/ha, 292 ml/ha	CO <sub>2</sub> Backpack
2016	Gainesville, TX	Dekalb, DKS 38-88	Monsanto	May 6	dryland	50, 125, 300, UTC	Sulfoxaflor, Flupyradifurone	70 g/ha, 292 ml/ha	CO <sub>2</sub> Backpack
2016	Monticello, AR A	Dekalb, DKS 38-88	Monsanto	Jun 16	furrow	50, 125, 300, UTC	Flupyradifurone	512 ml/ha	Self-propelled plot sprayer
2016	Monticello, AR B	Pioneer, 84P80	Pioneer	Jun 17	furrow	50, 125, 300, UTC	Flupyradifurone	512 ml/ha	Self-propelled plot sprayer
2016	Griffin, GA	Dekalb, DKS 53-53	Monsanto	Jun 22	sprinkler	50, 125, 300, 500, UTC	Flupyradifurone	292 ml/ha	CO <sub>2</sub> Backpack
2016	Florence, SC	Dekalb, DKS 38-88	Monsanto	May 5	dryland	50, 125, 300, UTC	Sulfoxaflor, Flupyradifurone	70 g/ha, 292 ml/ha	CO <sub>2</sub> Backpack

<sup>a</sup> Hybrids provided as a courtesy or purchased with the understanding that data would be analyzed to produce insecticide use guidance aggregated across hybrids. TAMU Sorghum Breeding Program seed courtesy of W. Rooney (Department of Soil and Crop Sciences, Texas A&M University), SSC seed from Southern States Cooperative.

<sup>b</sup> Naturally occurring aphid populations were allowed to colonize plots and increase to the targeted aphid population densities prior to use of either sulfoxaflor or flupyradifurone per label instructions. Targeted aphid densities varied by location and year. A spray was re-applied if aphid population growth again exceeded the targeted aphid population densities.

<sup>c</sup> Rates of 70 g/ha, 292 ml/ha, and 512 ml/ha correspond to 1.0 oz/ac, 4.0 fl oz/ac, and 7.0 fl oz/ac, respectively.

of infestation by sugarcane aphid. Aphid colonization occurred prior to boot stage of plant growth in all locations and years (Table 2-1). Naturally occurring aphid populations were allowed to colonize plots and increase to targeted aphid population densities prior to use of an insecticide. These targeted aphid densities varied by location and year, but at least four of the following were used: 0, 25, 50, 125, 250, 300, 430, 500 aphids per leaf, along with a non-insecticide check that was left unsprayed (Table 2-1). Plots were arranged in a randomized complete block of targeted aphid density treatments and replicated three (Rosenberg TX, 2015) or four (all other location-years) times. Once populations reached the targeted aphid density treatments, the plots of that treatment were treated with either sulfoxaflor (Transform WG, Dow AgroSciences, Indianapolis, IN) or flupyradifurone (Sivanto Prime, Bayer Crop Science, Monheim am Rhein, Germany). Plots were sprayed using either a CO<sub>2</sub> powered backpack sprayer, or a self-propelled plot sprayer. Spray tips varied by location. Label requirements were followed to ensure proper canopy penetration and coverage with a minimum final spray volume of 93.6 L per ha. The insecticide was re-applied if aphid population growth again exceeded the targeted aphid population densities. Sorghum hybrid and plot irrigation varied by location (Table 2-1) while all other agronomic management adhered to standard regional practices (e.g., Trostle and Fromme 2010).

### **2.2.2. Insect Sampling, Aphid Population Estimation, and Yield Evaluation**

In-season aphid measurements were taken weekly to every one to two weeks starting after first aphid detection. Data collection continued until two consecutive

observations of aphid decline were recorded. For Corpus Christi, TX, and Winnsboro, LA, in 2014 and 2015 and Rosenberg, TX, in 2015, average aphid density per plot was calculated using visual estimates of aphids on two leaves per plant and 10 randomly selected plants per plot. The leaves selected were the first healthy leaf (> 80% of the leaf was green) from the bottom of the plant and the most recent completely unfurled leaf below the flag leaf (designated as bottom and top leaves, respectively). Aphids were counted individually up to 10, then were placed in the following categories: 11 to 25 aphids (midpoint count of 18), 26 to 50 aphids (38 midpoint), 51 to 100 aphids (75 midpoint), 101 to 500 aphids (300 midpoint), 501 to 1,000 aphids (750 midpoint), and > 1,000 aphids per leaf (1,500 set as high maximum based on field observations in the region [Brewer et al. 2017]). Plot averages for the bottom leaf, top leaf, and the average across both leaves (combining all data) were calculated using the actual counts, midrange, and high maximum values. For all other location-years, aphids were counted up to 50 per leaf, after which the populations were estimated by 10s up to 250, 50s up to 500, and 100s thereafter. The same leaf selection method was used to sample five plants (10 total leaves) for the Griffin, GA, location, and 10 plants (20 total leaves) for all other locations. Using these averages, cumulative aphid days were calculated using the formula  $\sum[(x_i+x_{i-1})/2] \times (t_i-t_{i-1})$ , where  $(x_i+x_{i-1})/2$  was the aphid density  $x$  between progressive sampling periods  $i$ , and  $(t_i-t_{i-1})$  was the number of days  $t$  between sampling periods (Kieckhefer et al. 1995). Maximum aphid density was the highest plot average density observed across all sampling dates (Brewer et al. 2017).

At maturity, the middle two rows of plots were harvested either with a small plot combine, or by hand and processed through a thresher. Yields were adjusted to 14% moisture and were recorded on a kg per ha basis. To attribute yield reduction to sugarcane aphid, all non-target insects were controlled for the duration of experiments. The pests varied by location and year. Up to two selected insecticides were applied after head emergence at labelled rates to all plots to control sorghum midge, stink bugs, and headworms (Trostle and Fromme 2010). Insecticides used were methomyl (Lannate LV, DuPont, Wilmington, DE), chlorantraniliprole (Prevathon, Dupont, Wilmington, DE), beta-cyfluthrin (Baythroid XL, Bayer CropScience LP, Research Triangle Park, NC.), and zeta-cypermethrin (Mustang-Maxx, FMC, Philadelphia, PA).

### **2.2.3. Aphid Population Growth and Yield—Aphid Population Relationships**

Population growth rate and doubling time were analyzed with regression techniques. Weekly aphid estimate data from non-insecticide treated plots were used during the observation periods when aphid populations were increasing. The data were fit to natural log-linear regression (Freund and Littell 2000). Population growth regression analysis was performed separately for each location-year of the experiment. The regression slope was used as an estimator of the field-based aphid population growth rate  $r$  (slope from regression analysis). Population doubling time, DT, measured in days, was calculated using the formula  $DT = [\log_e(2)]/r$ . Daily rate of increase,  $\lambda$ , was calculated using the formula  $\lambda = e^r$  (DeLoach 1974, Akbar et al. 2010). Only location-years with significant ( $P < 0.05$ ) regressions were used to calculate the regression



coefficients and the derived aphid growth estimates. For guidance in estimating economic thresholds sensitive to aphid population growth, population growth regressions were first aggregated and evaluated for heterogeneity of slopes using PROC GLM, with location as the covariate (Freund and Littell 2000).

To evaluate the yield--aphid relationship, individual plot averages of yield and the two aphid population estimates (maximum aphid density and cumulative aphid days) were analyzed separately with linear regression, where yield for each plot was the dependent variable and the plot-average aphid population estimate was the independent variable (Freund and Littell 2000). In previous research and preliminary analyses of the data sets here, linear regression described the yield--aphid relationship as well or better than higher order regressions (Brewer et al. 2017). Regression analysis was performed separately for each location-year of the experiment. For guidance in estimating economic injury level, yield--aphid regressions were first aggregated and evaluated for heterogeneity of slopes using PROC GLM, with location as the covariate (Freund and Littell 2000).

#### **2.2.4. Economic Injury Levels and Thresholds**

The economic injury level (EIL) was calculated by the formula  $EIL = C/(V \cdot I \cdot D \cdot K)$ , where C = control cost, V = \$ value of grain, K is the proportion of the insect population controlled, and I\*D is loss per insect. I\*D was estimated as the slope of yield—maximum aphid density regression (Pedigo 1999). Fit of the regressions using maximum aphid density and aggregating the slopes across location-year regressions

were more consistent and as good as or better than the fit using yield—cumulative aphid-day regressions (see results). Therefore, maximum aphid density EILs were calculated, and the regression estimates for calculating cumulative aphid-day EILs were presented (Ragsdale et al. 2007). The slope value(s) used depended on the outcome of the slope heterogeneity analysis. For K, 0.95 was used as a conservative estimate although efficacy trials indicate greater control with commonly used products such as sulfoxaflor and flupyradifurone (Buntin and Roberts 2016). Economic thresholds (ET) were derived by the formula  $ET = EIL * \lambda^{-x}$ , where  $\lambda$  is the daily rate of increase, and  $x$  is time expressed as days of lag time needed to implement a management tactic (in this application, a foliar-applied insecticide) (Pedigo 1999). The value(s) of  $\lambda$  used depended on the outcome of the slope heterogeneity analysis.

The market values (V) and control costs (C) used in calculation of the EIL were taken from several sources. Stiles and Stark (2016) estimated that the application cost of using a personally owned, self-propelled sprayer was \$4.94/ha. Their costs included repairs, maintenance, depreciation, and interest. Custom application costs for ground application averaged \$16.99/ha (range from \$14.53 to \$19.50/ha) from 2015-2017 (Falconer et al. 2016, TACR 2016, Langemeier 2017). During that same period, custom aerial application costs averaged \$24.71/ha (range from \$15.69 to \$37.48/ha) (Falconer et al. 2016, TACR 2016, Langemeier 2017). We obtained retail prices of commonly used insecticides for sugarcane aphid control that included sulfoxaflor and flupyradifurone. We used total control costs, including insecticide and application, to provide three generalized cost estimates: 1) a low cost of \$24.71/ha (\$10.00/ac), which included the

lower priced insecticide applied at a standard rate (lower range of labeled rate) with a grower-owned sprayer, 2) a mid-range cost of \$37.06/ha (\$15.00/acre), representative of custom ground application of the lower priced insecticide, and 3) a high cost of \$49.42/ha (\$20.00/ac), which included the more expensive of the insecticides applied at a standard rate (lower range of labeled rate) by a custom aerial applicator. Market values used in the calculation of the economic injury level represented three possible grain sorghum prices for the southern U.S.A.: 1) a low value estimate of \$137.79/metric ton sorghum (\$3.50/bu), 2) a mid-range value of \$157.47/metric ton (\$4.00/bu), and 3) a high value of \$177.16/metric ton (\$4.50/bu). These sorghum price estimates were not a forecast, rather they represented a range of prices observed over the past two years (USSP 2018).

## **2.3. Results and Discussion**

### **2.3.1. Aphid Population Growth and Doubling Time**

Field population growth was adequately described by a simple linear function in the majority of the 15 location-years (Table 2-2). For 12 location-years, the linear model was significant at  $\alpha = 0.05$ . For these location-years, the slope point estimates, serving as a measure of  $r$ , varied from 0.079 to 0.193, and  $R^2$  varied from 0.138 to 0.822. Data from two other location-years showed poor fit to the linear regression model ( $P > 0.30$ ): Winnsboro, LA, in 2015, and Monticello B, AR, in 2016. These locations experienced tremendous population growth, followed by a sudden decline, resulting in no greater than two consecutive dates with increasing aphid populations. Another location (Griffin,

**Table 2-2 Sugarcane aphid field population growth rates calculated using data from regional field experiments on sorghum hybrids susceptible to sugarcane aphid in the southern U.S.A., 2014-2016; Reprinted with permission from Gordy et al. (2019)**

Year	Location	n	Population growth rate, $r^{ab}$	Intercept <sup>a</sup>	$R^2$	$P$ value	Daily rate of increase, $\lambda^c$	Doubling time, DT <sup>d</sup>
2014	Corpus Christi TX	16	0.087 [0.010,0.165]	4.646 [3.810,5.483]	0.297	0.0291	1.091	7.920
2014	Winnsboro LA	20	0.187 [0.144,0.230]	1.618 [0.914,2.321]	0.822	<0.0001	1.205	3.710
2015	Corpus Christi TX	64	0.102 [0.080,0.124]	0.886 [0.284,1.487]	0.582	<0.0001	1.107	6.800
2015	Winnsboro LA	16	0.078 [-0.099,0.254]	4.602 [3.855,5.350]	0.162	0.3232	---	---
2015	Rosenberg TX	12	0.092 [0.040,0.143]	5.175 [4.676,5.674]	0.609	0.0027	1.096	7.560
2015	Monticello AR	28	0.123 [0.008,0.238]	1.636 [-0.606,3.877]	0.138	0.0363	1.131	7.230
2015	Griffin GA	12	0.086 [-0.012,0.185]	3.332 [2.696,3.969]	0.275	0.0798	---	---
2016	Corpus Christi TX	20	0.095 [0.019,0.171]	1.817 [0.482,3.152]	0.289	0.0175	1.100	7.300
2016	Winnsboro LA	16	0.185 [0.057,0.313]	0.679 [-0.791,2.150]	0.408	0.0078	1.203	3.750
2016	Rosenberg TX	16	0.193 [0.114,0.272]	2.366 [1.471,3.260]	0.664	0.0001	1.213	3.590
2016	Gainesville TX	24	0.179 [0.141,0.216]	-0.224 [-1.013,0.564]	0.815	<0.0001	1.196	3.880
2016	Monticello AR A	12	0.079 [0.024,0.134]	4.968 [4.558,5.378]	0.510	0.0091	1.082	8.760
2016	Monticello AR B	12	0.012 [-0.079,0.100]	4.884 [4.212,5.556]	0.007	0.7986	---	---
2016	Griffin GA	16	0.129 [0.055,0.203]	4.299 [3.674,4.924]	0.500	0.0022	1.138	5.370
2016	Florence SC	20	0.102 [0.039,0.165]	2.710 [1.751,3.669]	0.389	0.0033	1.107	6.790
	Mean (All) <sup>ef</sup>		0.128	2.643	0.504		1.137	6.06
	High <sup>eg</sup>	92	0.128 [0.091,0.166]	2.157 [1.589,2.725]	0.341	<0.0001	1.137	6.06
	Low <sup>eg</sup>	171	0.046 [0.024,0.069]	3.249 [2.807,3.692]	0.093	<0.0001	1.048	14.93

<sup>a</sup> Parameter estimate followed by 95% confidence interval.

<sup>b</sup> Population growth rate in non-treated plots (slope of the log-linear regression).

<sup>c</sup> Daily rate of increase,  $\lambda=e^r$ .

<sup>d</sup> Population doubling time, in days,  $DT = [\log_e (2)]/r$ .

<sup>e</sup> Only parameters from significant regressions were used to calculate means. The three non-significant location-year regressions, Winnsboro, LA (2015), Griffin, GA (2015), and Monticello B, AR (2016) were excluded from pooled parameters because of lack of significance of individual regressions ( $P>0.05$ ).

<sup>f</sup> All indicates the arithmetic mean of point estimates for all significant regressions.

<sup>g</sup> High and Low indicates the common point estimates from regression analysis for the high and low growth rate groupings with slopes greater or lower than 0.127, respectively.

GA, in 2015), was marginally non-significant ( $P = 0.07$ ). These three location-years were excluded from calculating population parameters.

For the location-years with significant regressions, variation did not follow a pattern with regard to region or production practice. A more detailed assessment was conducted by combining all location-years and evaluating heterogeneity of slopes. Heterogeneity was detected (location by day interaction,  $F=1.94$ ; d.f =11, 262;  $P=0.0349$ ). I considered the arithmetic mean of the population growth rate regression coefficients (0.128) as a point of separation for two groups: a high population growth rate group (slope greater than 0.128 for 5 location-years) and a low population growth rate group (slope less than 0.128 for 7 location-years) (Table 2-2). Each group was then re-evaluated, and the slopes within the two groups were determined not to be different (heterogeneity of slopes was not detected,  $P>0.87$ ) (Freund and Littell 2000). The two estimated slopes of the high and low population growth rate groups resulted in daily rates of increase ( $\lambda$ ) of 1.137 and 1.048, respectively. The arithmetic mean for all 12 location-years with significant population growth regressions resulted in the same daily rate of increase as the regression of the high population growth rate group ( $\lambda=1.137$ ). Additionally, there was overlap of the 95% confidence intervals of the  $r$  point estimates. Therefore, the data were combined for these 12 location-years to obtain common point estimates for  $r$ , daily rate of increase,  $\lambda$ , and doubling time, DT. The common values across all 12 regressions were used for subsequent calculation of the economic thresholds. Values for individual locations were also presented for those interested in further study at specific localities (Table 2-2). Population growth rates of sugarcane

aphid observed in these field experiments were lower than those observed for greenbug on sorghum conducted under laboratory conditions (Kerns et al. 1989), but were similar to those observed for sugarcane aphid on sugarcane conducted under greenhouse conditions (Akbar et al. 2010).

### **2.3.2. Yield-aphid Relationships**

For maximum aphid density (MAD) and cumulative aphid day (CAD) yield regressions, 12 of 15 location years demonstrated a significant yield-aphid population linear regression. The three location-years where the regressions were not significant were Corpus Christi, Rosenberg, and Gainesville, TX, in 2016 ( $P>0.10$ , Table 2-3). This lack of significance can likely be attributed to a combination of factors. For the Corpus Christi location, maximum aphid density was relatively low, only exceeding 300 aphids per leaf in one plot. For Rosenberg and Gainesville locations, maximum aphid density levels above 250 aphids per leaf in untreated plots were sustained for less than 7 and 10 days, respectively. Additionally, there was ample soil moisture at these locations which likely reduced sugarcane aphid-induced plant stress in grain sorghum (J. Gordy, pers. obs.) and as previously documented for selected other cereal aphids (Oswald and Brewer 1997, Brewer et al. 2019). These regressions were not used for further analyses.

When combining the 12 significant regressions, heterogeneity of slopes was detected (location by aphid population interaction,  $F=2.97$ ; d.f =11, 225;  $P=0.0011$ ). For the maximum aphid density measure, the arithmetic mean of yield--aphid regression coefficients (-3.8) was used as a point of separation of the 12 location-years into two

groups: a high response/environmental susceptibility to sugarcane aphid group (slope less than -3.8 for six location-years) and a low response/environmental susceptibility to sugarcane aphid group (slope greater than -3.8, for six location-years) (Table 2-3). Each group was then re-evaluated and regression coefficients with each of the two groups were not significantly different (heterogeneity of slopes was not detected,  $P > 0.20$ ) (Freund and Littell 2000). The common slope for the maximum aphid density regression for high response/environmental susceptibility was  $-6.810 \pm 0.867$ , with an  $R^2$  of 0.362 ( $F=61.73$ ; d.f.=1, 109;  $P < 0.0001$ ). The common slope for the low response/environmental susceptibility group was  $-2.463 \pm 0.441$ , with an  $R^2$  of 0.216 ( $F=31.14$ ; d.f. = 1, 113;  $P < 0.0001$ ) (Table 2-3).

For cumulative aphid days, heterogeneity of slopes was detected when combining all significant regressions ( $F=12.34$ ; d.f =11, 225;  $P < 0.001$ ). Using the same procedure of creating two groups using the arithmetic mean of individual slopes as the demarcation of the groups (Table 2-2), heterogeneity of slopes was likely ( $P < 0.08$  for the two analyses); therefore, slope estimates of all significant regressions were presented. The model fit and variation of the parameters estimated for analyses of the maximum aphid density and cumulative aphid day data were similar as judged by the  $R^2$  values and 95% confidence intervals of the parameter estimates, but the slope grouping process was beneficial in reducing slope heterogeneity only for the maximum aphid density measurement. Also, standard aphid monitoring activities report mean values of measurements on individual dates for IPM decision-making (Johnston and Bishop 1987, Ragsdale et al. 2007, Kerns et al. 2015); therefore, we used maximum aphid density to

calculate two sets of economic injury levels and economic thresholds to help simplify IPM decision-making on susceptible hybrids.

### **2.3.3. Economic Injury Level and Economic Threshold**

Using three control costs and market values, a range of economic injury levels was calculated for the common slope -6.81 of the combined yield--maximum aphid density regressions, representing a relatively high response/environmental susceptibility to sugarcane aphid (Table 2-4). Using the same method, a range of economic injury levels was calculated for the common slopes -2.46 of the combined yield--maximum aphid density regressions, representing a relatively low response/environmental susceptibility to sugarcane aphid (Table 2-4). For the relatively high response/susceptibility group, economic injury level varied from 22 aphids per leaf when the grain market value was high and the cost of control was low, and 55 aphids per leaf when the grain value was low and cost of control was high (Table 2-4). Using mean daily rate of increase  $\lambda$  of 1.137, the economic threshold was calculated to be 32, 28, 25, and 19 aphids per leaf for 1, 2, 3, and 5 days lag time, respectively (Table 2-4). This range fell below the lower recommendation of 50 aphids per leaf proposed by Knutson et al. (2016). For the relatively low response/susceptibility group, economic injury level varied from 60 aphids per leaf when the grain market value was high and the cost of control was low, to 153 aphids per leaf when the grain value was low and cost of control was high (Table 2-4). Using mean daily rate of increase  $\lambda$  of 1.137, the calculated



**Table 2-3 Yield--aphid linear regressions for two aphid population estimates (maximum aphid density and cumulative aphid day) calculated using data from regional field experiments on sorghum hybrids susceptible to sugarcane aphid in the southern U.S.A., 2014-2016; Reprinted with permission from Gordy et al. (2019)**

Year	Location	n	Maximum Aphid Density				Cumulative Aphid Days			
			Intercept <sup>a</sup>	Slope <sup>ab</sup>	R <sup>2</sup>	P value	Intercept <sup>a</sup>	Slope <sup>a</sup>	R <sup>2</sup>	P value
2014	Corpus Christi TX	20	3240 [2916,3564]	-1.912 [-2.493,-1.221]	0.7264	<0.0001	3147 [2898,3395]	-0.162 [-0.201,-0.123]	0.8086	<0.0001
2014	Winnsboro LA	20	4345 [3736,4955]	-2.702 [-3.830,-1.570]	0.5847	<0.0002	4610 [4093,5127]	-0.270 [-0.35,-0.190]	0.7352	<0.0002
2015	Corpus Christi TX	16	2091 [1535,2647]	-2.543 [-4.512,-0.574]	0.3541	0.015	2054 [1521,2586]	-0.121 [-0.215,-0.027]	0.3535	0.0151
2015	Winnsboro LA	19	5974 [5083,6864]	-3.984 [-7.469,-0.499]	0.2549	0.0275	6045 [5405,6686]	-0.451 [-0.696,-0.206]	0.4704	0.0012
2015	Rosenberg TX	15	1425 [1042,1808]	-1.728 [-2.434,-1.022]	0.6827	<0.0001	1474 [1259,1690]	-0.086 [-0.104,-0.067]	0.8864	<0.0001
2015	Monticello AR	28	5014 [4481,5548]	-3.518 [-5.039,-1.997]	0.465	<0.0001	4923 [4372,5473]	-0.025 [-0.037,-0.013]	0.4074	0.0003
2015	Griffin GA	24	3225 [2665,3785]	-4.597 [-6.266,-2.927]	0.597	<0.0001	3072 [2600,3544]	-0.320 [-0.425,-0.216]	0.6472	<0.0001
2016	Corpus Christi TX	16	1755 [1012,2498]	-4.512 [-10.099,1.075]	0.1765	0.1052	1853 [993,2713]	-0.022 [-0.050,0.006]	0.1677	0.1152
2016	Winnsboro LA	16	5905 [5542,6269]	-4.673 [-6.692,-2.654]	0.6378	0.0002	5898 [5567,6230]	-0.445 [-0.619,-0.271]	0.6816	<0.0001
2016	Rosenberg TX	16	3459 [3002,3916]	-1.143 [-2.868,0.582]	0.1261	0.1772	3408 [3037,3778]	-0.005 [-0.011,0.001]	0.1413	0.1513
2016	Gainesville TX	16	1559 [771,2348]	0.406 [-0.917,1.729]	0.0301	0.5208	1735 [866,2604]	0.000 [-0.010,0.011]	0.0003	0.9483
2016	Monticello AR A	16	4040 [2666,5415]	-4.112 [-8.117,-0.107]	0.2572	0.0449	4358 [3701,5015]	-0.493 [-0.665,-0.321]	0.7302	<0.0001
2016	Monticello AR B	16	5047 [4282,5812]	-6.005 [-9.156,-2.853]	0.544	0.0011	4928 [4436,5420]	-0.449 [-0.601,-0.296]	0.7395	<0.0001
2016	Griffin GA	20	3056 [2204,3908]	-6.298 [-8.832,-3.764]	0.6023	<0.0001	3045 [2384,3707]	-0.526 [-0.686,-0.367]	0.7267	<0.0001
2016	Florence SC	16	2442 [1756,3128]	-3.708 [-6.353,-1.062]	0.3923	0.0094	2654 [2136,3171]	-0.293 [-0.417,-0.169]	0.6465	0.0002
	All Locations <sup>c</sup>	276	3553 [3272,3834]	-2.901 [-3.646,-2.155]	0.1775	<0.0001	2847 [2617,3077]	-0.0085 [-0.016,-0.001]	0.0171	0.0306
	High <sup>cd</sup>	111	4881 [4378,5385]	-6.810 [-8.528,-5.092]	0.3616	<0.0001	4744 [4337,5152]	-0.560 [-0.673,-0.447]	0.4744	<0.0001
	Low <sup>cd</sup>	115	3312 [2927,3697]	-2.463 [-3.338,-1.589]	0.2161	<0.0001	2320 [1985,2685]	0.011 [-0.003,0.026]	0.0207	0.1272

<sup>a</sup> Parameter estimate followed by 95% confidence interval.

<sup>b</sup> Means are included here for reference. For economic injury level and economic threshold calculations, the mean slope for maximum aphid density–yield regression was used as the point of separation to aggregate data as described in text.

<sup>c</sup> Only parameters from significant regressions were used to calculate means. The three non-significant location-year regressions, Corpus Christi, TX (2016), Rosenberg, TX (2016), and Gainesville, TX (2016), were excluded from pooled parameters because of lack of significance of individual regressions ( $P>0.05$ ).

<sup>d</sup> High and Low indicates the common point estimates from the regression analysis for the relatively high and low response/environmental susceptibility to sugarcane aphid groupings with slopes lower or greater than -3.815, respectively.

economic threshold increased to 88, 78, 70, and 54 aphids per leaf for 1, 2, 3, and 5 days lag time, respectively (Table 2-4). Some susceptible hybrids appear to be able to avoid yield loss when aphids exceed the suggested range of ETs (Table 2-4) if there is sufficient soil moisture and aphid populations build up quickly and rapidly decline as seen at the Corpus Christi, Rosenberg, and Gainesville, TX, locations in 2016.

Additionally, when field monitoring for aphids, caution should be taken to avoid errors in estimation based on sampling effort. Bowling et al. (2016) recommended sampling an upper and lower canopy leaf from 10 plants (20 total leaves) to calculate an estimate of aphids per leaf. A demonstration project in south Texas revised the procedure to 20 plants, 40 total leaves (Deleon et al. 2017). Additional analyses of large sugarcane aphid distribution and density data sets are needed to optimize the field sampling processes used to implement the economic thresholds presented here (Elliott et al. 2017) to consider the trade-off between precision and cost in terms of time associated with whole plant counts or reduced sampling (McCornack et al. 2008). Binomial and sequential sampling are two alternatives to the aphid density-based approach presented here. Giles et al. (2003) validated the effectiveness of a binomial, sequential sampling plan for greenbug in winter and spring wheat. Hodgson et al. (2004) recommended use of a tally threshold to calculate percent plants infested above a set soybean aphid density. Similar sampling methods for sugarcane aphid in grain sorghum may increase efficiency of scouting while maintaining an acceptable level of estimation while reducing the time and effort of scouting (Pedigo and Buntin 1993).

**Table 2-4 Economic injury levels (EIL) and economic thresholds (ET) calculated as sugarcane aphid per leaf, based on control cost, market price, and lag time for management application for yield—maximum aphid density regression of relatively high and low response/environmental susceptibility to sugarcane aphid groupings with slopes lower or greater than -3.815, respectively; Reprinted with permission from Gordy et al. (2019)**

Cost of Control <sup>a</sup> (\$/ha)	Market Price <sup>b</sup> (\$/metric ton)	EIL (aphids/leaf)	ET with different lag times (days) <sup>c</sup>			
			1	2	3	5
-----Low Response/Environmental Susceptibility-----						
24.71	137.79	77	66	59	53	41
	157.47	67	58	51	46	36
	177.16	60	52	46	41	32
37.06	137.79	115	99	88	79	61
	157.47	101	87	77	69	53
	177.16	90	77	68	61	47
49.42	137.79	153	132	117	105	81
	157.47	134	116	103	92	71
	177.16	119	103	91	82	63
Mean		102	88	78	70	54
-----High Response/Environmental Susceptibility-----						
24.71	137.79	28	24	21	19	15
	157.47	24	21	19	17	13
	177.16	22	19	16	15	11
37.06	137.79	42	36	32	29	22
	157.47	36	31	28	25	19
	177.16	32	28	25	22	17
49.42	137.79	55	48	42	38	29
	157.47	49	42	37	33	26
	177.16	43	37	33	30	23
Mean		37	32	28	25	19

<sup>a</sup> Corresponds to \$10, \$15, and \$20 per acre, range based on information from Falconer et al. (2016), Stiles and Stark (2016), TACR (2016), and Langemeier (2017).

<sup>b</sup> Corresponds to \$3.50, \$4.00, and \$4.50/bushel, range based on information from USSP (2018).

<sup>c</sup> Lag time are the days anticipated before a sugarcane aphid control tactic is implemented, to avoid populations reaching the economic injury level.

In addition to adoption of thresholds for determining necessity and timing of chemical control, Quisenberry and Pears (1998) and Michels and Burd (2007) included use of resistant cultivars or hybrids as part of the IPM approach for managing Russian wheat aphid and greenbug, respectively. Revisions to thresholds proposed here will likely be needed for sorghum hybrids partially resistant to sugarcane aphid (Armstrong et al. 2015), as found for other cereal aphids (Randolph et al. 2003). Also, natural enemies are common in this system and may affect aphid population growth (Brewer et al. 2017). Therefore, consideration for beneficial insects, both predators and parasitoids, and subsequent adjustments in using the proposed economic thresholds are worthy of additional study, as done by Giles et al. (2003) and Hoffmann et al. (1991).

Based on data across 15 location-years, field-based population growth rates of sugarcane aphids were calculated and a range of economic thresholds based on maximum aphid density during infestation of vegetative growth were estimated. This research included a wide range of geography, environmental conditions, production practices, cropping seasons, and sugarcane aphid population ranges. Using this data set, economic thresholds ranged from 19 to 132 aphids per leaf, with mean economic injury levels of 37 aphids per leaf for environments where aphid populations grow relatively rapidly and 102 aphids per leaf for environments where populations grow relatively slowly. The threshold range presented here overlaps with the range of 50 to 125 aphids per leaf previously suggested by Knutson et al. (2016). Thresholds of 50 to 100 aphids per leaf have been implemented in south Texas (Deleon et al. 2017), while the low estimate of 50 aphids per leaf has been implemented in the fast growing populations

recently experienced in the southern High Plains (Szczepaniec 2018). Modifications of these thresholds are appropriate based on changes in commodity price, management costs, and desired outcomes of their respective sorghum pest management program. However, without site-specific knowledge of what regulates slow- or fast-growing aphid populations and given cost and market price variability of the system, a 40 aphid per leaf threshold is most prudent to use across the range of hybrid, environmental, and market conditions experienced in this study.

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### 3. TALLY-BASED THRESHOLDS AS AN ALTERNATIVE TO DENSITY-BASED THRESHOLDS FOR SUGARCANE APHID, *Melanaphis sacchari*, (HEMIPTERA: APHIDIDAE) IN GRAIN SORGHUM

#### 3.1. Introduction

Sugarcane aphid, *Melanaphis sacchari* Zehntner (Hemiptera: Aphididae), has become a major pest of grain sorghum, *Sorghum bicolor* (L.) Moench, in North America since outbreaks were first detected on the crop in Texas and surrounding states in 2013. Sugarcane aphid occurred on sugarcane in Florida as early as 1977 and in Louisiana in 1999. It was reported that sugarcane aphid would feed on *Sorghum spp.*, but it was not considered a significant sorghum pest until this recent outbreak (Mead 1978, Denmark 1988, White et al. 2001, Bowling et al. 2016). In 2015, sugarcane aphid populations confirmed in the major sorghum producing regions of the United States (Bowling, et al. 2016, Brewer et al. 2017). This new population found on sorghum has been reported to exhibit low genetic diversity and consists of a dominant clonal lineage, which was likely introduced into the Americas from either Africa or Asia (Nibouche et al. 2018, Harris-Schultz et al. 2017).

Sugarcane aphid is well suited to tropical and subtropical environments (Akbar 2010, Singh et al. 2004), and has very high population growth potential on cultivated sorghum grown in North America (Gordy et al. 2019). Sugarcane aphids feed primarily on the underside of leaves, removing large amounts of plant fluids and exuding honeydew, which supports growth of sooty mold. The feeding and sooty mold

negatively impact resource availability and disrupts the photosynthetic capability of the plant. Infested plants can exhibit stress symptoms including stunting, yellowing leaves, die-back, and incomplete or no panicle emergence (Bowling et al. 2016). Infestations during sorghum vegetative growth through grain development can cause significant yield loss (Gordy et al. 2019).

As applied to management of sugarcane aphid on grain sorghum, the economic injury level is the lowest sugarcane aphid population level where economic loss of grain yield equals control costs. The economic threshold is the population level below the economic injury level that prompts use of an insecticide or alternative management tactic to prevent the population from exceeding the economic injury level (Pedigo 1999). A density-based threshold is supported by direct counts of aphids, which can be time-consuming. Accurate classification of densities above or below a density-based threshold is critical to its use, but the classification may be sensitive to a few unusual observations (i.e., very high individual counts and variation in counting among samplers) (Thomas et al. 2018). A tally threshold is defined as the number of individuals needed to be present for a sampling unit to be considered infested (Capinera 2008). A proportion-tally threshold approach (i.e. proportion of infested sampling units exceeding the tally threshold) is more resistant to the effects of unusual observations, particularly at low density levels (Pedigo and Buntin 1994). Hall et al. (2007) demonstrated that rust mite, *Phyllocoptruta oleivora* Ashmead, densities can be estimated using the proportion of samples infested rather than actual mite counts, and others have shown use of tally

thresholds can improve decision-making in both field and greenhouse applications (Rogers et al. 1994, Naranjo et al. 1996, Lee et al. 2005).

For pest management decision-making, the objective is to determine if the pest population is above or below a critical level such as the economic threshold (Pedigo and Buntin 1994). Gordy et al. (2019) established a regionally-applicable density-based economic threshold of 40 aphids per leaf for sugarcane aphid on grain sorghum considering data from a wide range of growing conditions, grain values, and control costs. Decision-making for sugarcane aphid management using insecticides may be performed more quickly and efficiently using a tally-based threshold if it is a reasonable substitute for a density-based threshold. Recently, Lindenmayer et al. (2020a) developed a binomial sequential sampling plan for sugarcane aphid and investigated a broad array of tally thresholds to substitute for the density-based thresholds proposed by Gordy et al. (2019). The objectives here were to further explore the empirical relationship between aphid density and a tally threshold proportion by evaluating the infestation proportion – aphid density relationship using linear and polynomial regression. Validation and time efficiency in a farm setting was considered using a separate data set, specifically comparing several proposed tally-based thresholds derived from the empirical relationships to the previously established density-based threshold approach in decision-making.

## **3.2. Materials and Methods**

### **3.2.1. Experimental Design and Manipulation**

Sugarcane aphid abundance data were collected from 11 field experiments conducted at three locations in Texas from 2015-2018 (Table 3-1). Experiments consisted of large on-farm trials and small plot replicated experiments using both sugarcane aphid-susceptible and partially resistant hybrids, except two locations in 2015 where only a susceptible hybrid was evaluated. A mid- to late-planting time (early April through June, depending on geography), based on local standards, was used to maximize chances of infestation by sugarcane aphid. Aphid colonization occurred prior to boot stage of plant growth in all locations and years.

Sugarcane aphid densities varied across locations, years, and sorghum hybrids. In addition, densities were also manipulated by insecticides applied when populations reached a given level. Several experimental designs were used: a randomized complete block of targeted aphid density treatments, a split plot with insecticide treatment as main plots and sorghum hybrid as subplots, and replicated strip plots of different hybrids. Individual plot sizes ranged from 9.14 m by four rows (small plots of randomized complete block and split plot designs) to 290 m by six rows (strip plots). For all experiments, there were three (Rosenberg TX, 2015) or four (all other location-years) replications (Table 3-1). Naturally occurring aphid populations were allowed to colonize plots and increase, or were manipulated with an insecticide in order to achieve a range of aphid densities. These targeted aphid densities varied by location and year, but at least four of the following were chosen for each experiment in 2015 and 2016: 50, 125, 250,

**Table 3-1 Location, sorghum hybrids, and sampling effort in Texas (2015-2018) used to generate proportion – mean density regression parameters for selected tally thresholds**

Year	Location	Hybrid Type(s) <sup>a</sup>	Sampling Dates	Samples Used	Plot Type <sup>b</sup>	Aphid Densities <sup>c</sup>
2015	Corpus Christi, TX	S	9	180	RCB	Targeted densities
2015	Rosenberg, TX	S	11	165	RCB	Targeted densities
2016	Corpus Christi, TX	S, R	12	960	RCB	Targeted densities
2016	Rosenberg, TX	S, R	7	576	RCB	Targeted densities
2016	Gainesville, TX	S, R	10	960	RCB	Targeted densities
2017	Corpus Christi, TX	S, R	9	717	Split	Near zero, unmanaged
2017	Rosenberg, TX	S, R	8	640	Split	Near zero, unmanaged
2017	Gainesville, TX	S, R	5	392	Strip	Unmanaged
2018	Rosenberg, TX(A)	S, R	8	512	Split	Near zero, unmanaged
2018	Rosenberg, TX(B)	S, R	3	288	Strip	Unmanaged
2018	Gainesville, TX	S, R	2	192	Strip	Unmanaged

<sup>a</sup> S=aphid susceptible, R=partially aphid resistant (See chapter 4)

<sup>b</sup> RCB = randomized complete block; Split = split plot; Strip = strip plots.

<sup>c</sup> All plots were initially colonized by natural populations of sugarcane aphid and left unmanaged (unmanaged), were treated with insecticide at detection of *M. sacchari* (near zero), or treated with an insecticide when *M. sacchari* reached several targeted densities to obtain a range of infestations.



300, 500 aphids per leaf. Split plot experiments in 2017 and 2018 had hybrids as main plots with subplots that were sprayed with insecticide (when aphids were detected) or left unsprayed. The replicated strip plot designs were left unsprayed.

For randomized complete block and split plot designs, aphids were managed by applying sulfoxaflor (Transform WG, Corteva Agriscience, Indianapolis, IN) or flupyradifurone (Sivanto Prime, Bayer Crop Science, Monheim am Rhein, Germany) at labeled rates. Treated plots were sprayed using a CO<sub>2</sub>-powered backpack sprayer or self-propelled plot sprayer. Spray tips varied by location, and a minimum final spray volume of 93.6 L per ha was used. Insecticides were re-applied if aphid population growth again exceeded the targeted aphid population densities. Agronomic management practices adhered to standard regional practices (e.g., Trostle and Fromme 2010).

### **3.2.2. Insect Sampling, Aphid Population Estimation, and Yield Evaluation**

For all experimental designs, plot sections where aphids were sampled ranged in size from 9.14 to 12.19 m by four rows, with row spacing of 0.76 to 1.02 m. In-season aphid densities were estimated weekly (small plots of the randomized complete block and split plot designs) or every two to three weeks (strip plots) after first aphid detection. Twenty leaves from 10 plants were sampled per plot, then an aphid per leaf average was calculated for the plot. The leaves selected were the lowest green leaf of the plant and the most recent completely unfurled leaf below the flag leaf. For Corpus Christi and Rosenberg locations in 2015, estimates of aphids on each leaf were made visually per Gordy et al. (2019). For all other location-years, aphids were counted to 50 per leaf, after

which the populations were estimated by 10s up to 250, 50s up to 500, and 100s thereafter. Sampling was stopped after detection of aphid population decline for consecutive sampling periods. This sampling protocol was used to generate the previously established density-based economic threshold (Gordy et al. 2019).

### **3.2.3. Infestation Proportion –Aphid Density Regression**

Previous work established a density-based economic threshold of 40 aphids per leaf for susceptible hybrids across a range of environments. Simple linear regression and regression comparison were done to estimate yield loss per aphid per leaf (Gordy et al. 2019). Also considering aphid population growth rate, grain price, and cost of control (Gordy et al. 2019), the density based economic threshold was calculated (Pedigo 1999). Tally thresholds of 25, 50 or 100 aphids per leaf were of interest because tally thresholds have been proposed in the Extension literature (Knutson et al. 2016) and in research efforts on use of multiple tactics for management (Haar et al. 2019). Lindenmayer et al. (2020a) proposed a binomial sequential sampling approach that considered an array of tally thresholds. To further explore the infestation proportion – aphid density relationship for the selected 25, 50, and 100 tally thresholds consider practical by these authors, individual plot data of aphid densities and the infestation proportions (proportion of leaves with more than 25, 50, or 100 aphids per leaf) were analyzed using linear and polynomial (second order) regression (Freund and Littell 2000).

Regression analysis was performed separately for each location-year to test for model significance ( $p < 0.05$ ). For linear regression, infestation proportion was the

dependent variable and the mean aphid density was the independent variable as previously done by Lindenmayer 2020a). Mean aphid density squared was added as a second independent variable for the second order polynomial regression model. Next, heterogeneity of slopes was evaluated across the 11 location-years (Freund and Littell 2000), as done by Gordy et al. (2019) when proposing a regionally-applicable density-based economic threshold. Threshold proportions for aphid density tallies (>25, >50, and >100 aphids per leaf) were calculated based on the economic threshold of 40 aphids per leaf, identified by Gordy et al. (2019). The tally threshold infestation proportion was defined as the proportion of leaves (using equal number of upper and lower leaves) with more than 25, 50, or 100 aphids (tally threshold) equivalent to an average density of 40 aphids per leaf predicted by the parameters from the infestation proportion – aphid density regression models.

#### **3.2.4. Field Validation**

In 2018, five commercial grain sorghum fields in the upper Texas Gulf Coast region (one of the three original areas of data collection) were sampled to compare decisions made based on the original density-based threshold of 40 aphids per leaf and several proposed tally-based thresholds. In each field, aphids were counted on the lowest green leaf and the uppermost leaf below the flag leaf on 50 randomly selected plants spread over approximately a half hectare (19 total field samples independent of data used for generating the original model). A fixed sample size of 100 leaves (two leaves from 50 plants) was used, which exceeded recommendations by Knutson et al. (2018) to

evaluate at least 10 plants in four field locations for sugarcane aphid sampling and exceeded past sampling effort of sugarcane aphid in small plot research (Brewer et al. 2017). It was also consistent with a recommendation by Elliot et al. (2017) and Lindenmayer et al. (2020b) of using the standard uppermost and lowermost leaf sample unit combination (completely unfurled leaf with >80% green tissue) as the preferred method for assessing sugarcane aphid density to avoid estimation problems associated with within-plant variability. For the 100 leaves inspected, proportions of leaves with >25, >50, and >100 aphids per leaf and mean aphid density were recorded. Using the parameters from the linear and polynomial regression models from the original dataset, expected infestation proportion was calculated for the field observed mean aphid density, resulting in six expected infestation proportions (three tally thresholds by two regression models) used in the validation exercise. For each tally threshold, expected infestation proportion was regressed on the field observed infestation proportion. The slope of the expected and observed infestation proportion was estimated, and the hypothesis of equality was tested (slope = 1) (PROC REG, SAS Institute 2014).

The degree of agreement in decision-making when using proposed equivalent tally threshold proportion and mean density approaches was also evaluated using the independent field data. Each plot was assigned a treatment decision based on mean density (apply an insecticide if mean density was >40 aphids per leaf) and tally threshold proportion (apply an insecticide if observed infestation proportion was greater than calculated proportion utilizing regression parameters). This exercise was performed using the three proposed tally threshold proportions estimated from the two infestation

proportion – aphid density regression models (linear and polynomial). For each of these six models, the number of times where insecticide use decisions differed were recorded and percent agreement of decision was calculated by dividing the number of occasions where decisions agreed by the total number of field samples. The time required to conduct both threshold approaches (density-based threshold and tally-based threshold) was recorded. A paired t-test was performed to compare time efficiency of aphid density estimation and tally threshold approaches (SAS Institute 2014).

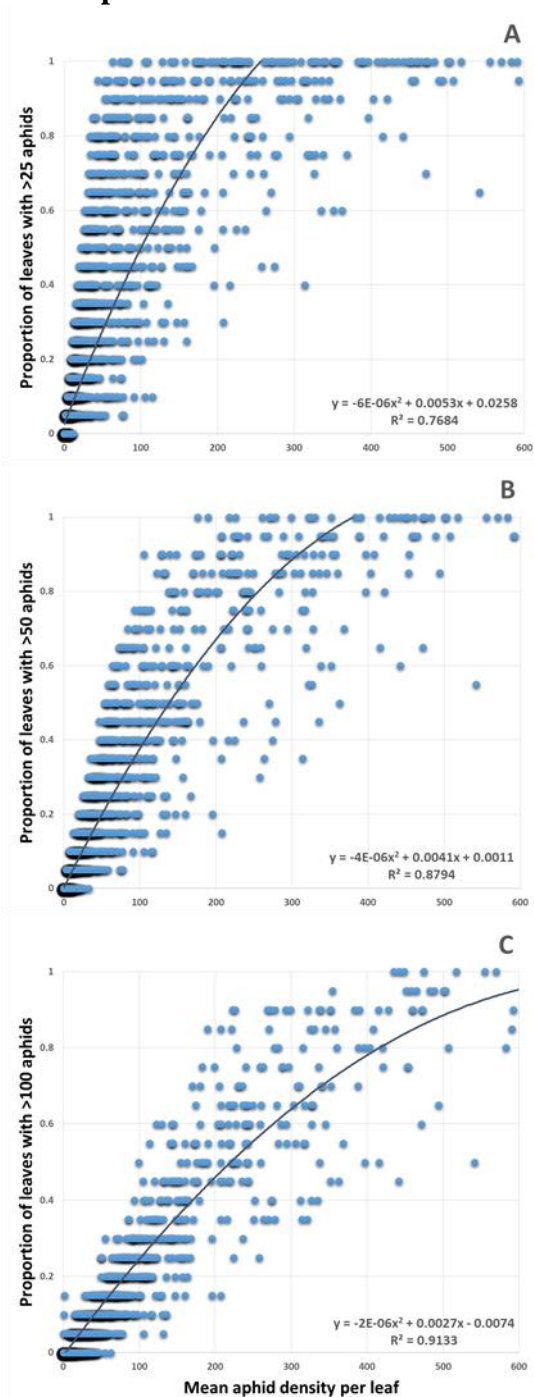
### **3.3. Results and Discussion**

#### **3.3.1. Infestation Proportion –Aphid Density Regression**

All 33 individual linear regressions for the three proportion tally thresholds and location-years were significant at  $\alpha=0.05$  (individual regressions provided in supplemental Table 7-1). As mean aphid density approached and exceeded 200 aphids per leaf, the density-proportion relationship reached an asymptote of a proportion of 1.0 (all leaves infested with aphids at or above the tally threshold) which could not be exceeded, as illustrated in scatter plots of data aggregated across the location-years (Figure 3-1). This issue was most extreme at the lowest tally threshold of 25 aphids per leaf. The data were fit to a second order polynomial regression in an effort to better describe the relationship. The 33 individual polynomial regressions were significant and better characterized the relationship as determined by an increase in  $R^2$  values (individual regressions provided in supplemental Table 7-2).

Many of the infestation proportion – aphid density regressions appeared similar within the three tally thresholds and across location-years. Inspections for slope heterogeneity included possible groupings among years, locations, and aphid-susceptible and resistant hybrids as done by Gordy et al. (2019). No distinct groups could be identified; therefore, parameters from linear and polynomial regressions using aggregated data were used to estimate the three tally threshold proportions (n=5582 for each tally threshold). When all location-years were aggregated,  $R^2$  values for the polynomial regression models were 0.77, 0.88, and 0.91 for the tally thresholds of >25, >50, and >100 aphids per leaf, respectively (Figure 3-1, Table 3-2). These were improved from the results of the linear regressions using the aggregated data with  $R^2$  values of 0.59, 0.75, and 0.86 for the same tally thresholds (Table 3-2). Using the density-based threshold of 40 aphids per leaf, a tally threshold proportion was calculated for each tally threshold. For the tally thresholds of >25, >50, and >100 aphids per leaf, the tally threshold proportion was 0.23, 0.16, and 0.11, respectively, when using parameters from the polynomial regression aggregating all location-years (Table 3-2). Using a fixed sample size of 100 leaves divided evenly between the top and bottom half of the plant, 23, 16, and 10 leaves with more than 25, 50, and 100 aphids per leaf, respectively, would be equivalent to an economic threshold of 40 aphids per leaf. Using a wide array of tally thresholds and linear regression, Lindenmayer et al. (2020a) reported that a 20% infestation of leaf pairs with more than 100 aphids (equal to a tally threshold of 50 aphids per leaf) was equivalent to a density of 37.5 aphids per leaf. Our results using polynomial regression are similar: a 0.16 tally threshold proportion (16%)

**Figure 3-1 Polynomial regression models for observed proportion – mean density regressions for >25 (A), >50 (B), and >100 (C) aphid per leaf tally thresholds for field experiments in Texas 2015-2018**



**Table 3-2 Infestation proportion – mean density regression parameters for tally thresholds of >25, >50, and >100 aphids per leaf, combined across 11 location-years in Texas, 2015-2018**

Regression	Tally Threshold	Slope $\alpha^a$	Slope $b^a$	Intercept <sup>a</sup>	$R^2$	Threshold Proportion <sup>b</sup>
Polynomial	>25 aphids/leaf	$-5.9e^{-6} \pm 9.0e^{-8}$	$0.0053 \pm 4.7e^{-5}$	$0.0258 \pm 0.0017$	0.7684	0.2284
	>50 aphids/leaf	$-4.0e^{-6} \pm 5.0e^{-8}$	$0.0041 \pm 2.7e^{-5}$	$0.0010 \pm 0.001$	0.8794	0.1586
	>100 aphids/leaf	$-1.9e^{-6} \pm 3.0e^{-8}$	$0.0027 \pm 1.7e^{-5}$	$0.0010 \pm 0.001$	0.9133	0.1060
Linear	>25 aphids/leaf	--	$0.0026 \pm 2.9e^{-5}$	$0.0542 \pm 0.002$	0.5927	0.1583
	>50 aphids/leaf	--	$0.0023 \pm 1.8e^{-5}$	$0.0204 \pm 0.001$	0.7492	0.1124
	>100 aphids/leaf	--	$0.0019 \pm 9.9e^{-6}$	$0.0015 \pm 0.001$	0.8636	0.0759

<sup>a</sup> Parameter estimate from regressions where  $y=ax^2+bx+\text{intercept}$  (polynomial, second order) and  $y=bx+\text{intercept}$  (linear), followed by the standard error. The model output is the incremental increase of the proportion of leaves with >X aphids (25, 50, or 100) for each aphid present on a leaf.

<sup>b</sup> Tally threshold proportion as defined as the proportion of leaves with >X aphids (tally threshold), which is equivalent to a mean aphid density of 40 aphids per leaf (the previously established density-based threshold [Gordy et al. 2019]).

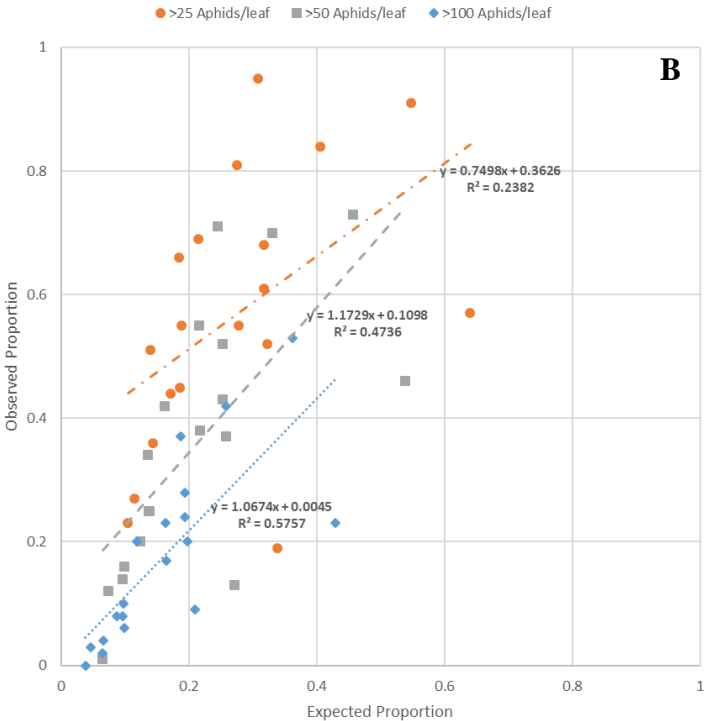
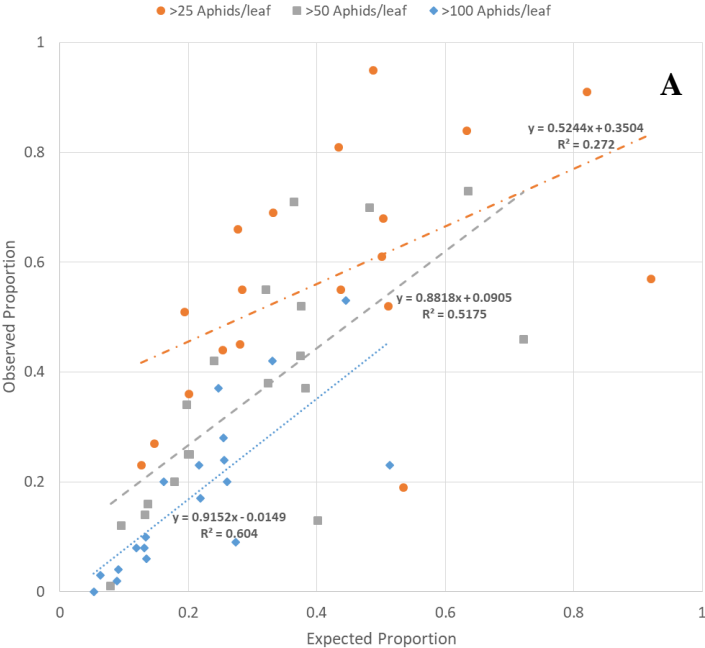


of leaves infested with >50 aphids per leaf (tally threshold) is equivalent to the density-based thresholds of 40 aphids per leaf proposed by Gordy et al. (2017). The similarity with the nominal tally thresholds suggested by Knutson et al. (2016) further supports use of tally-based thresholds in a farm setting.

### **3.3.2. Field Validation for Decision-Making**

Moving to a farm setting, independent validation and comparison of time efficiencies was conducted using 19 samples from commercial grain sorghum fields. A *F*-test was performed to test if the slope of the regression of observed values of the proportion of infested plants (dependent variable) on expected values (predicted proportion of infested plants based on regression parameters, independent variable) was equal to 1, for both the linear and polynomial regression parameters. For the linear regression parameters, slope of the observed on expected values for tally thresholds of >25, >50, and >100 aphids per leaf were not different from 1 ( $F=0.59$ ; d.f.=1,17;  $p=0.4523$ ,  $F=0.33$ ; d.f.=1,17;  $p=0.5718$ , and  $F=0.09$ ; d.f.=1,17;  $p=0.7653$ , respectively). For the polynomial regression parameters, the *F*-test indicated a significant difference for the tally threshold of >25 aphids per leaf ( $F=5.22$ ; d.f.=1,17;  $p=0.0354$ ). In contrast, tally thresholds of >50 and >100 aphids per leaf were not different from a slope of 1 ( $F=0.33$ ; d.f.=1,17;  $p=0.5745$ , and  $F=0.22$ ; d.f.=1,17;  $p=0.6429$ , respectively) (Figure 3-2). These results support that use of >50 and >100 aphid per leaf tally threshold models should provide best predictions of the tally threshold proportion across the range of infestation proportions observed in the

**Figure 3-2 Regression models for observed versus expected proportions for polynomial (A) and linear (B) models from independent validation data from commercial grain sorghum fields in Texas in 2018**



validation data set. This is in agreement with the higher  $R^2$  values of the regression equations with using the >50 and >100 aphid tally thresholds (Table 3-2). When comparing agreement of the insecticide use decision between the density- and tally-based threshold methods for the independent data set, the tally-threshold proportions calculated using the linear regression-derived parameters resulted in 79%, 84%, and 94% agreement for the >25, >50, and >100 aphid/leaf threshold proportions, respectively. For the polynomial parameter-derived tally-threshold proportions, all three tally thresholds demonstrated 100% agreement (19 of 19) with the density-based threshold approach. A high level of confidence in consistency of treatment decision-making for insecticide use was seen when using the tally threshold proportions of 0.16 and 0.11 for the >50 and >100 aphid tally thresholds, respectively, obtained from the polynomial models (Table 3-2).

In side-by-side sampling using a fixed sample size of 100 leaves (50 plants, sampling upper and lower leaf) for both the density-based threshold and tally-based threshold methods, the tally threshold approach required approximately half the time to assess 100 leaves (mean of 8 min., 32 sec.,  $n=19$ ) compared to the aphid density threshold approach (mean of 16 min., 41 sec.,  $n=19$ ) ( $t= 9.52$ ; d.f. = 18;  $p < 0.001$ ).

Others have found similar success utilizing tally thresholds. For example, Hodgson et al. (2004) evaluated tally thresholds of 20 and 40 soybean aphids per leaf and concluded that the >40 aphid per leaf tally threshold with an action threshold (tally threshold proportion) of 0.84 (proportion of leaves with >40 aphids) accurately reflected an economic threshold of 250 aphids per leaf. Similar work has been done on whitefly in

cotton and rust mite in citrus where tally thresholds of three whitefly per leaf and zero or two rust mites per  $\text{cm}^2$  were suggested for use in decision-making, respectively, although tally threshold proportions were not specifically given (Naranjo et al. 1996, Hall et al. 2007). Additionally, Ward et al. (1985) demonstrated that density-based thresholds and tally-based threshold proportions yielded similar accuracy at low to intermediate aphid densities when a fixed sample size of 100 tillers was used to evaluate cereal aphids on wheat.

### **3.4. Conclusions**

Research here and consensus with others (Knutson et al. 2016, Lindenmayer et al. 2020a) supports use of an infestation proportion of a tally threshold for decision-making on use of insecticides to manage sugarcane aphid on grain sorghum (i.e., a tally-based threshold). By evaluating the infestation proportion – aphid density relationship using polynomial regression, the best estimates for tally-based thresholds were an infestation proportion of 0.16 for a tally threshold of >50 aphids per leaf or 0.11 for a tally threshold of >100 aphids per leaf. Both served as a suitable alternative to the density-based threshold proposed by Gordy et al. (2019) based on similarity with the nominal tally thresholds suggested by Knutson et al. (2016) and the development of binomial sequential sampling plans for sugarcane aphid that utilized a wide array of tally thresholds (Lindenmayer et al. 2020a). Moving to a farm setting, independent validation further supported use of tally thresholds of >50 and >100 aphids per leaf, and a comparison of time efficiencies indicated the tally threshold approach required

approximately half the time to complete a 100-leaf sample, compared to density-based threshold approach of Gordy et al. (2017). For those interested in additional efficiencies when using tally thresholds in a sampling program, sequential sampling procedures using tally thresholds (Lindenmayer et al. 2020a) may further reduce the sampling effort of the standard 100 leaves used in our validation exercise. Last, the example tally-based infestation proportions given here are a substitute for the density-based threshold of 40 aphids per leaf that was considered an average suitable across a range of environmental conditions and sorghum hybrids susceptible to sugarcane aphid (Gordy et al. 2019). The regression parameters in Table 3-2 may be used by a pest manager to estimate a tally threshold infestation proportion for adjustments made to the density-based economic threshold should more detailed information be available for a region or sorghum hybrid.

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## 4. FIELD ASSESSMENT OF APHID DOUBLING TIME AND YIELD OF SORGHUM SUSCEPTIBLE AND PARTIALLY RESISTANT TO SUGARCANE

APHID, *Melanaphis sacchari* (HEMIPTERA: APHIDIDAE)

### 4.1. Introduction

Sugarcane aphid, *Melanaphis sacchari* Zehntner (Hemiptera: Aphididae), has an expansive world distribution, including Africa, Asia, Australia, and parts of Central and South America, and is a pest of grain sorghum, *Sorghum bicolor* (L.) Moench, in many locations (Singh et al. 2004). While sugarcane aphid was previously reported to feed on *Sorghum* spp. (Denmark 1988), it was not considered a significant sorghum pest in North America until the outbreaks on sorghum beginning in 2013 (Bowling et al. 2016). Within three years, confirmed sugarcane aphid populations on sorghum were reported in all major sorghum growing areas of the southern U.S., Mexico, and Caribbean Islands (Bowling et al. 2016, Harris-Schultz et al. 2017).

Grain sorghum is an important crop in North America. From 2012 to 2019, between 1.9 and 3.2 million hectares of grain sorghum were harvested annually in the U.S. and production in Mexico is extensive (USDA-NASS 2020, Bowling et al. 2016). Recently, Gordy et al. (2019) calculated a range of economic injury levels and estimated a broadly applicable density-based economic threshold of 40 aphids per leaf by assessing the yield-aphid density relationship of aphid susceptible sorghum hybrids across a range of geographies, aphid pressure, and environmental conditions. Additionally, Lindenmayer et al. (2020) developed a sequential sampling plan for sugarcane aphid in

grain sorghum based on this economic threshold. However, neither approach considered adjustment based on resistance of grain sorghum against the aphid.

Cereal crops resistant to aphids have been deployed in the U.S. for some time in response to aphid invaders, including various cultivars resistant to Russian wheat aphid, *Diuraphis noxia* Kurdjumov, and greenbug, *Schizaphis graminum* Rondani (Quisenberry and Peairs 1998, Michels and Burd 2007, Brewer et al. 2019). Since the outbreak of sugarcane aphid on sorghum in the U.S., screening of sorghum breeding lines and hybrids have been conducted under greenhouse conditions and on selected crop stages, by both private and public researchers (e.g., Armstrong et al. 2017, Peterson et al. 2018). These efforts have led to the commercialization of many sorghum hybrids with varying degrees of apparent partial resistance to sugarcane aphid, as reported in peer-reviewed and trade literature (Haar et al. 2019, USCP 2020).

To evaluate susceptibility of grain sorghum hybrids purported to be partially resistant to sugarcane aphid in a full-season field setting, and to provide guidance on need for supplemental insecticide use when growing these hybrids, field evaluations of the relationship between grain yield and aphid population estimates were conducted across numerous hybrids and a range of environmental conditions and repeated across several years and locations. These data were used to analyze aphid population growth rates and field susceptibility of aphid-resistant hybrids compared to known aphid-susceptible hybrids.

## **4.2. Methods and Materials**

### **4.2.1. Experimental Design and Manipulation**

Field experiments were conducted at various locations (Texas, Louisiana, Arkansas, and South Carolina) across the southern U.S. in 2016, 2017, and 2018 (Table 4-1). Known aphid-susceptible hybrids and purported sugarcane aphid-resistant hybrids were planted using several experimental designs: a randomized complete block of different aphid density treatments manipulated with an insecticide, a split plot with insecticide treatment as main plots and sorghum hybrid as subplots, and replicated strip plots. The 18 purported partially resistant and seven susceptible hybrids used in the experiments were either known to be or expected to be well adapted to commercial production in areas represented by research locations (Table 4-2). Randomized complete block and split plot experiments consisted of plots with lengths ranging from 9.14 to 12.19 m by four rows spaced 0.76 to 1.02 m apart. Strip plots ranged in length from 60 to 290 meters and were four or six rows wide with the same row spacing. There were four replications in all experiments. Considering local production practices, a mid- to late-planting time was used to maximize chances of natural colonization by sugarcane aphid. Natural infestations of sugarcane aphid occurred during vegetative growth stages in all locations and years.

Sugarcane aphid densities varied across locations, years, sorghum hybrids, and as manipulated with insecticide in selected experiments detailed as follows. In the randomized complete block experiments, an insecticide was used when 50, 125, and 300 aphids per leaf were detected in separate treatments, along with a non-insecticide check

that was left unsprayed (Table 4-1). Split plot experiments in 2017 and 2018 consisted of main plots that were either sprayed with an insecticide when aphids were detected or were left unsprayed. Strip plot experiments were planted and aphids were left unmanaged with regard to insecticide use. For randomized complete block and split plot designs, aphids were managed by applying sulfoxaflor (Transform WG, Corteva Agriscience, Indianapolis, IN) or flupyradifurone (Sivanto Prime, Bayer Crop Science, Monheim am Rhein, Germany) at labeled rates. Insecticide-treated plots were sprayed using a self-propelled plot sprayer or CO<sub>2</sub> powered backpack sprayer. A minimum final spray volume of 93.6 L per ha was used with spray tips varying by location. The insecticide was re-applied if aphid population growth again exceeded the targeted aphid population densities. Agronomic management practices and management of other insect pests adhered to standard regional practices (e.g., Knutson et al. 2018, Trostle and Fromme 2010). To attribute yield reduction to sugarcane aphid, non-target pest insects were controlled. The pests varied by location and year. Up to two selected insecticides were applied after head emergence at labelled rates to all plots to control sorghum midge, stink bugs, and headworms (Knutson et al. 2018). Insecticides used were methomyl (Lannate LV, DuPont, Wilmington, DE), chlorantraniliprole (Prevathon, Dupont, Wilmington, DE), beta-cyfluthrin (Baythroid XL, Bayer CropScience LP, Research Triangle Park, NC.), and zeta-cypermethrin (Mustang-Maxx, FMC, Philadelphia, PA) and were selected based on their lack of activity on sugarcane aphid.

#### **4.2.2. Insect Sampling, Aphid Population Estimation, and Yield Evaluation**

The three years, six locations, and three field plot designs resulted in a wide range of environmental conditions and aphid population growth. In-season aphid measurements were taken weekly (randomized complete block and split plot) to every two to three weeks (strip plots) starting after first aphid detection. In each plot, the lowest green leaf and the upper-most unfurled leaf below the flag leaf were examined on 10 randomly selected plants, for a total of 20 leaves per plot (Brewer et al. 2017, Gordy et al. 2019). Aphids were counted up to 50 per leaf, after which the populations were estimated by 10s up to 250, 50s up to 500, and 100s thereafter. Sampling was stopped after two weeks of aphid population decline. The total number of aphids per plot was divided by the 20 leaves sampled to estimate the average aphid density per leaf for each plot, which was used for aphid population growth estimation. Maximum aphid density was defined as the highest average plot density per leaf observed across all sampling dates for each hybrid location-year. This measure was used for estimating the yield-aphid density relationship. In previous research, maximum aphid density described aphid density-yield relationships as well or better than cumulative aphid days (Gordy et al. 2019).

At maturity, the middle two rows of plots (0.0020 to 0.0026 hectare) for the randomized complete block and split plot experiments, or 0.0004 hectare for the strip plot experiments were harvested. Harvest was performed with a small-plot combine (randomized complete block and split-plot design) or by hand and processed through a

**Table 4-1 Experimental details for regional field experiments on sorghum hybrids susceptible and partially resistant to sugarcane aphid in the southern U.S., 2016-2018**

Year	Location	Experimental Code	Irrigation	# Hybrids <sup>a</sup>	Plot Type	Aphid Manipulation <sup>b</sup>
2016	Corpus Christi, TX	CC16	dryland	6	Randomized Complete Block	50, 125, 300, unmanaged
2016	Winnsboro, LA	LA16	furrow	3	Randomized Complete Block	50, 125, 300, unmanaged
2016	Rosenberg, TX	RB16	dryland	6	Randomized Complete Block	50, 125, 300, unmanaged
2016	Gainesville, TX	GV16	dryland	6	Randomized Complete Block	50, 125, 300, unmanaged
2016	Monticello, AR	AR16	furrow	4	Randomized Complete Block	50, 125, 300, unmanaged
2016	Florence, SC	SC16	dryland	2	Randomized Complete Block	50, 125, 300, unmanaged
2017	Corpus Christi TX	CC17	dryland	10	Split plot	near zero, unmanaged
2017	Gainesville TX	GV17	dryland	7	Strip plot	unmanaged
2017	Rosenberg TX	RB17	dryland	9	Split plot	near zero, unmanaged
2018	Rosenberg TX A	RB18A	dryland	8	Split plot	near zero, unmanaged
2018	Rosenberg TX B	RB18B	dryland	8	Strip plot	unmanaged

<sup>a</sup> See Tale 4-2 for specific hybrids used in each experiment as identified by the experimental code.

<sup>b</sup> Naturally occurring aphid populations were allowed to colonize plots and increase to the targeted aphid population densities prior to use of either sulfoxaflor or flupyradifurone per label instructions. A spray was re-applied if aphid population growth again exceeded the targeted aphid population densities. Unmanaged indicates no insecticide was applied to control aphids, near zero indicates insecticides were applied as soon as aphids were detected to keep aphid populations near zero.

**Table 4-2 Sorghum hybrids used in field trials in the southern U.S., 2016-2018. Hybrids were described by seed company or supplier as partially resistant or susceptible to *Melanaphis sacchari***

Hybrid (abbreviation)	Source <sup>a</sup>	Brand	Resistant/Susceptible <sup>b</sup>	Maturity	Experimental Code
2752/430 (TX430)	TAMU	N/A	Susceptible	Medium	CC17, RB17
2752/2783 (TX2783)	TAMU	N/A	Partially Resistant	Medium	CC17, RB17
625Y (W625Y)	Warner Seeds Inc.	Warner	Partially Resistant	Medium	GV17
7051 (W7051)	Warner Seeds Inc.	Warner	Partially Resistant	Medium-full	CC17, RB17
74GB17 (74GB17)	Dyna-Gro Seed	Dyna-Gro	Partially Resistant	Medium-full	CC17, GV17, RB18A
83P17 (83P17)	Corteva (DuPont)	Pioneer	Partially Resistant	Medium-full	AR16
83P56 (83P56)	Corteva (Dupont)	Pioneer	Partially Resistant	Medium-full	CC17, GV17, RB17, RB18A, RB18B
84P80 (84P80)	Corteva (Dupont)	Pioneer	Susceptible	Medium-full	AR16
844E (W844E)	Warner Seeds Inc.	Warner	Partially Resistant	Medium-full	CC17
ADV G3247 (G3247)	Advanta	Alta Seeds	Partially Resistant	Medium-full	RB18A, RB18B
BH 3822 (BH3822)	BH Genetics	BH Genetics	Susceptible	Medium-early	GV17
BH 4100 (BH4100)	BH Genetics	BH Genetics	Partially Resistant	Medium-full	CC16, CC17, GV16, GV17, RB16, RB17
Chr0L0242 (CHR242)	Chromatin Inc.	Chromatin	Partially Resistant	Medium-full	GV17
DKS 37-07 (DK3707)	Bayer (Monsanto)	Dekalb	Partially Resistant	Medium-early	CC16, GV16, GV17, SC16, RB16, LA16
DKS 38-88 (DK3888)	Bayer (Monsanto)	Dekalb	Susceptible	Medium-early	CC16, GV16, SC16, AR16, RB16, LA16
DKS 45-23 (DK4523)	Bayer (Monsanto)	Dekalb	Susceptible	Medium	RB18A, RB18B
DKS 47-07 (DK4707)	Bayer (Monsanto)	Dekalb	Partially Resistant	Medium	RB18A, RB18B
DKS 48-07 (DK4807)	Bayer (Monsanto)	Dekalb	Partially Resistant	Medium	CC16, GV16, RB16, RB17
DKS 53-67 (DK5367)	Bayer (Monsanto)	Dekalb	Susceptible	Medium-full	CC16, CC17, GV17, RB16, RB17, RB18A, RB18B
EXP 481 (481)	Bayer (Monsanto)	Dekalb	Partially Resistant	Medium-full	RB18A, RB18B
EXP E44 (WE44)	Warner Seeds Inc.	Warner	Partially Resistant	Medium	CC17
REV9782 (REV9782)	Terral Seed	Rev	Susceptible	Medium-full	LA16
SP73B12 (73B12)	S&W Seed	Sorghum Partners	Partially Resistant	Medium-full	CC17, GV17
SP7715 (SP7715)	S&W Seed	Sorghum Partners	Partially Resistant	Medium-full	CC16, CC17, GV16, RB16, RB17
SP78M30 (78M30)	S&W Seed	Sorghum Partners	Partially Resistant	Medium-full	RB18A, RB18B

<sup>a</sup> Hybrids provided as a courtesy or purchased with the understanding that data would be analyzed to produce insecticide use guidance aggregated across hybrids. TAMU Sorghum Breeding Program seed courtesy of W. Rooney (Department of Soil and Crop Sciences, Texas A&M University).

<sup>b</sup> As described by respective seed companies or seed source.



thresher (strip plots). Yields were adjusted to 14% moisture and were recorded on a kg per ha basis.

#### **4.2.3. Aphid Population Growth and Yield—Aphid Population Relationships**

Aphid population growth rate and doubling time were estimated with regression techniques. Weekly aphid density estimates of increasing populations were used from non-insecticide treated plots. The data were fit to natural log-linear regression (Freund and Littell 2000). Population growth regression analysis was performed separately for each hybrid and location-year of the experiment, and additional analyses were conducted for significant regressions ( $P < 0.05$ ). To evaluate if a single combined regression for susceptible or partially resistant hybrids was representative of all location-years, population growth data was aggregated across all location-year combinations and evaluated for heterogeneity of slopes using PROC GLM, with location as the covariate (Freund and Littell 2000). If slope heterogeneity was detected and no groupings across locations or years helped explain the heterogeneity, then the slope parameter resulting from the regression analysis for each hybrid by location-year combination was used as an estimator of the field-based aphid population growth rate  $r$ . Population doubling time, DT, measured in days, was calculated using the formula  $DT = [\log_e(2)]/r$ . Daily rate of increase,  $\lambda$ , was calculated using the formula  $\lambda = e^r$  (DeLoach 1974, Gordy et al. 2019).

To evaluate the yield-aphid density relationship, individual yields and the maximum aphid density for each plot were analyzed using linear regression, where yield for each plot (measured in kg per ha) was the dependent variable and the maximum

aphid density (measured in aphids per leaf) was the independent variable (Freund and Littell 2000). In previous research, linear regression described the yield-aphid density relationship as well or better than higher order regressions (Gordy et al. 2019, Brewer et al. 2017). Following the same procedure for the population growth analyses, regression analysis was performed separately for each hybrid and location-year of the experiment. For each hybrid, location-year combinations demonstrating a significant yield-aphid density relationship were then aggregated and evaluated for heterogeneity of slopes using PROC GLM, with location as the covariate (Freund and Littell 2000). If slope heterogeneity was detected and no groupings across locations or years helped explain the heterogeneity, then the regression estimates resulting from the regression analysis for each hybrid by location-year combination was reported. If no slope heterogeneity was detected across location-years, then the common regression estimates were reported.

### **4.3. Results**

#### **4.3.1. Aphid Population Growth and Doubling Time**

Field population growth was adequately described by a simple log-linear function in the majority of hybrids across the 10 location-years evaluated (Table 4-3). For 58 of the 61 hybrids and location-years, the linear model was significant at  $\alpha = 0.05$ . For susceptible hybrids, the slope point estimates, serving as a measure of the field observed population growth rate  $r$ , varied from 0.031 to 0.346 (mean=0.154,  $n=16$ ), and  $R^2$  varied from 0.133 to 0.978. For purported resistant hybrids,  $r$  varied from 0.017 to 0.304 (mean=0.092,  $n=42$ ) and  $R^2$  varied from 0.119 to 0.940. The three hybrids that

showed poor fit to the linear model ( $P > 0.60$ ) were all from the same location-year (Monticello AR, 2016). This location-year experienced very high aphid population growth, followed by a sudden decline, resulting in no greater than two consecutive dates with increasing aphid populations in three of the four hybrids that were evaluated. Two additional location-years (Rosenberg TX B, 2018 and Gainesville TX 2018) were not included in the analysis as there were no hybrids for which aphid populations were sustained for more than two data collection dates.

For the location-years with significant regressions, heterogeneity of slopes was detected for both susceptible (location by date interaction,  $F=13.20$ ; d.f =9, 359;  $P<0.0001$ ) and resistant (location by date interaction,  $F=13.32$ ; d.f =8, 1019;  $P<0.0001$ ) hybrids. Variation in aphid population growth for each hybrid did not follow a pattern with regard to region, year, or production practice. Specifically, grouping data across locations or years for each hybrid did not provide identifiable homogenous groups (i.e., slope heterogeneity was still detected when re-running the analysis for these subgroups [analyses not shown]). Therefore,  $r$ , DT, and  $\lambda$  was reported separately (Table 4-3). The range of the population growth rate for known susceptible hybrids was 0.031 to 0.304 (mean = 0.154,  $n = 16$ ) and for the purported partially aphid-resistant hybrids the range was 0.017 to 0.304 (mean = 0.092,  $n = 42$ ). Susceptible hybrids demonstrated population doubling times of less than half on average of that of purported resistant hybrids: DT range for known susceptible hybrids was 2.0 to 22.4 (mean = 6.2 days,  $n = 16$ ) and for the purported partially aphid-resistant hybrids the DT range was 2.3 to 40.8 (mean = 13.4 days,  $n = 42$ ) (Table 4-3).

These differences reflected a wide range of maximum aphid densities observed: from 6 to 352 aphids per leaf for partially resistant hybrids and from 67 to 1025 aphids per leaf for known susceptible hybrids (Table 4-4). For example, population growth was slow and maximum aphid density was low at Corpus Christi TX in 2016 and at Gainesville TX in 2017, while the Rosenberg TX location in 2016 experienced high aphid population growth and maximum aphid density. During the 12 days when populations were increasing at the Rosenberg TX location in 2016, population doubling time ranged from 2.0-2.15 days for susceptible hybrids to 2.6-2.84 for resistant hybrids, an increase of 21-39%.

#### **4.3.2. Yield-Aphid Density Relationships and Decision-Making**

For the known susceptible hybrids, seven of twenty hybrid and location-year combinations demonstrated a significant yield-aphid density linear regression (Table 4-4). Four of these hybrid location-year regressions were included with other experiments in Gordy et al. (2019) to estimate a range of economic injury levels for susceptible hybrids. When performing a regression of combined data of these seven hybrids, heterogeneity of slopes was not detected (location by aphid population interaction across all susceptible hybrids,  $F=0.39$ ; d.f.=6, 87;  $P=0.8826$ ); therefore a common slope was used in a validation of the previously calculated range of economic injury levels. The common slope for the combined regression of the seven hybrids was  $-5.341 \pm 0.758$ , with an  $R^2$  of 0.366 ( $F=49.61$ ; d.f.=1, 87;  $P<0.0001$ ) (Table 4-4). The slope of the yield–

**Table 4-3 Sugarcane aphid field population growth rates from regional field experiments on partially resistant and susceptible sorghum hybrids in the southern U.S., 2016-2018**

Year	Location	Hybrid, R/S	n	Population growth rate, $r^{ab}$		Intercept <sup>a</sup>		$R^2$	$P$ value	Daily rate of increase, $\lambda^c$	Doubling time, DT <sup>d</sup>
2016	Corpus Christi TX	SP7715,R	24	0.023	[0.009,0.037]	-0.721	[-1.462,0.019]	0.3350	0.0030	1.023	30.14
		BH4100,R	24	0.034	[0.006,0.062]	-1.087	[-2.552,0.378]	0.2256	0.0190	1.035	20.39
		DK3888,S	24	0.108	[0.081,0.136]	-3.793	[-5.246,-2.341]	0.7477	<0.0001	1.114	6.42
		DK3707,R	24	0.017	[0.004,0.029]	-0.389	[-1.056,0.278]	0.2455	0.0138	1.017	40.77
		DK4807,R	24	0.020	[0.004,0.037]	-0.570	[-1.444,0.303]	0.2239	0.0195	1.020	34.66
		DK5367,S	24	0.077	[0.046,0.108]	-2.491	[-4.097,-0.885]	0.5488	<0.0001	1.080	9.00
2016	Winnsboro LA	DK3707,R	16	0.076	[0.011,0.142]	0.747	[-0.009,1.503]	0.3069	0.0260	1.079	9.12
		DK3888,S	16	0.157	[0.073,0.240]	1.346	[0.387,1.204]	0.5379	0.0012	1.170	4.41
		REV9782,S	16	0.163	[0.053,0.274]	1.152	[-0.119,2.424]	0.4182	0.0068	1.177	4.25
2016	Rosenberg TX	BH4100,R	12	0.251	[0.206,0.295]	1.796	[1.441,2.152]	0.9406	<0.0001	1.285	2.76
		DK3707,R	12	0.267	[0.214,0.319]	1.497	[1.076,1.918]	0.9276	<0.0001	1.306	2.60
		DK3888,S	12	0.346	[0.289,0.404]	1.575	[1.114,2.036]	0.9475	<0.0001	1.413	2.00
		DK4807,R	12	0.256	[0.187,0.325]	1.125	[0.571,1.678]	0.8721	<0.0001	1.292	2.71
		DK5367,S	12	0.323	[0.289,0.356]	1.898	[1.629,2.167]	0.9788	<0.0001	1.381	2.15
		SP7715,R	12	0.244	[0.166,0.322]	1.887	[1.261,2.512]	0.8297	<0.0001	1.276	2.84
2016	Gainesville TX	DK3707,R	20	0.115	[0.076,0.154]	-1.673	[-3.068,-0.279]	0.6814	<0.0001	1.122	6.03
		DK3888,S	20	0.167	[0.123,0.210]	-1.754	[-3.12,-0.197]	0.7828	<0.0001	1.182	4.15
		DK4807,R	20	0.139	[0.113,0.166]	-2.840	[-3.78,-1.902]	0.8744	<0.0001	1.149	4.99
		DK5367,S	20	0.190	[0.156,0.225]	-2.896	[-4.124,-1.667]	0.8834	<0.0001	1.209	3.65
		SP7715,R	20	0.110	[0.080,0.140]	-1.796	[-2.874,-0.718]	0.7663	<0.0001	1.116	6.30
		BH4100,R	20	0.167	[0.123,0.210]	-1.754	[-3.312,-0.197]	0.7828	<0.0001	1.182	4.15
2016	Monticello AR	DK3707,R	12	0.015	[-0.055,0.086]	3.651	[3.123,4.180]	0.0231	0.6373	-	-
		DK3888,S	12	0.079	[0.024,0.133]	4.974	[4.566,5.383]	0.5100	0.0091	1.082	8.77
		83P17,R	12	-0.010	[-0.092,0.071]	3.981	[3.368,4.593]	0.0078	0.7846	-	-
		84P80,S	12	0.011	[-0.078,0.099]	4.892	[4.225,5.560]	0.0069	0.7969	-	-
2016	Florence SC	DK3707,R	16	0.124	[0.064,0.185]	1.997	[1.010,2.324]	0.5802	0.0006	1.132	5.59
		DK3888,S	16	0.160	[0.085,0.236]	2.450	[1.629,3.272]	0.5953	0.0005	1.174	4.33
2017	Corpus Christi TX	BH4100,R	24	0.036	[0.020,0.052]	-0.731	[-1.377,-0.084]	0.4958	0.0001	1.037	19.25
		DK5367,S	24	0.174	[0.083,0.131]	-2.487	[-3.442,-1.531]	0.8022	<0.001	1.113	6.45
		83P56,R	24	0.092	[0.070,0.113]	-2.217	[-3.077,-1.357]	0.7846	<0.0001	1.096	7.53
		TX430,S	24	0.130	[0.099,0.161]	-3.230	[-4.475,-1.986]	0.7775	<0.0001	1.139	5.33
		TX2783,R	24	0.058	[0.035,0.080]	-1.396	[-2.311,-0.480]	0.7634	0.0002	1.060	11.95
		SP73B12,R	24	0.042	[0.027,0.060]	-0.948	[-1.612,-0.284]	0.5758	<0.0001	1.043	16.50

**Table 4-3, continued**

Year	Location	Hybrid, R/S	n	Population growth rate, $r^{ab}$	Intercept <sup>a</sup>	$R^2$	$P$ value	Daily rate of increase, $\lambda^c$	Doubling time, $DT^d$
2017	Corpus Christi TX	SP7715,R	18	0.042 [0.032,0.053]	-0.921 [-1.365,-0.482]	0.8112	<0.0001	1.043	16.50
		W7051,R	24	0.034 [0.025,0.044]	-0.879 [-1.258,-0.499]	0.7228	<0.0001	1.035	20.39
		W844E,R	18	0.066 [0.037,0.096]	-1.502 [-2.703,-0.302]	0.5846	0.0002	1.068	10.50
		WE44,R	24	0.056 [0.040,0.072]	-1.404 [-2.054,-0.753]	0.7043	<0.0001	1.058	12.38
2017	Gainesville TX	CHR242,R	48	0.304 [0.015,0.045]	1.432 [1.085,1.779]	0.2662	0.0002	1.355	2.28
		DK3707,R	48	0.023 [0.004,0.041]	1.707 [1.279,2.135]	0.1187	0.0165	1.023	30.14
		BH3822,S	48	0.031 [0.008,0.054]	3.235 [2.688,3.782]	0.1335	0.0107	1.031	22.36
		BH4100,R	48	0.019 [0.006,0.032]	0.959 [0.660,1.259]	0.1646	0.0042	1.019	36.48
		W625Y,R	48	0.035 [0.018,0.052]	1.727 [1.326,2.127]	0.2655	0.0002	1.036	19.80
		SP73B12,R	48	0.023 [0.010,0.036]	1.264 [0.963,1.565]	0.2163	0.0009	1.023	30.14
		83P56,R	48	0.059 [0.039,0.079]	2.262 [1.800,2.724]	0.4335	<0.0001	1.061	11.75
		TX2783,R	24	0.094 [0.066,0.122]	-0.033 [-0.693,0.627]	0.6856	<0.0001	1.099	7.37
2017	Rosenberg TX	BH4100,R	24	0.042 [0.015,0.070]	0.336 [-0.315,0.987]	0.3131	0.0045	1.043	16.50
		TX430,S	24	0.094 [0.074,0.115]	0.256 [-0.222,0.734]	0.8064	<0.0001	1.099	7.37
		DK4807,R	24	0.067 [0.037,0.097]	0.322 [-0.380,1.023]	0.4924	0.0001	1.069	10.35
		DK5367,S	24	0.134 [0.110,0.158]	-0.044 [-0.608,0.521]	0.8584	<0.0001	1.143	5.17
		W7051,R	24	0.064 [0.039,0.088]	-0.081 [-0.663,0.501]	0.5606	<0.0001	1.066	10.83
		SP7715,R	24	0.033 [0.007,0.058]	0.411 [-0.190,1.011]	0.2383	0.0155	1.034	21.00
		83P56,R	24	0.052 [0.015,0.089]	0.582 [-0.281,1.446]	0.2823	0.0076	1.053	13.33
		W844E,R	24	0.054 [0.025,0.082]	0.304 [-0.360,0.967]	0.4115	0.0007	1.055	12.84
		DK3707,R	20	0.158 [0.098,0.218]	0.682 [-0.363,7.727]	0.6280	<0.0001	1.171	4.39
		DK3816,S	20	0.082 [0.052,0.113]	0.272 [-0.257,0.801]	0.6420	<0.0001	1.085	8.45
		DK4523,S	20	0.082 [0.065,0.099]	-0.016 [-0.607,0.275]	0.8549	<0.0001	1.085	8.45
		DK4707,R	20	0.195 [0.152,0.238]	-0.118 [0.864,0.628]	0.8359	<0.0001	1.215	3.55
2018	Rosenberg TX A	74GB17,R	20	0.104 [0.080,0.128]	0.153 [-0.260,0.566]	0.8248	<0.0001	1.110	6.66
		SP78M30,	20	0.142 [0.117,0.167]	0.211 [-0.226,0.648]	0.8867	<0.0001	1.153	4.88
		83P56,R	20	0.061 [0.031,0.092]	0.301 [-0.226,0.828]	0.5025	0.0005	1.063	11.36
		3247,R	20	0.100 [0.079,0.121]	0.192 [0.178,0.563]	0.8441	<0.0001	1.105	6.93
		Mean, Resistant <sup>e</sup>		0.092				1.10	13.4
		Mean, susceptible <sup>e</sup>		0.154				1.17	6.2

<sup>a</sup> Parameter estimate followed by 95% confidence interval.

<sup>b</sup> Population growth rate in non-treated plots (slope of the log-linear regression).

<sup>c</sup> Daily rate of increase,  $\lambda=e^r$ .

<sup>d</sup> Population doubling time, in days,  $DT = [\log_e(2)]/r$ .

<sup>e</sup> Arithmetic mean across all significant regressions for resistant or susceptible hybrids.

\*Rosenberg TX B and Gainesville TX 2018 locations were not included because there were only two sampling dates for each trial location.

aphid density relationship for the seven hybrids observed here was similar to that reported by Gordy et al. (2019) for susceptible hybrids under high response/environmental conditions where aphid reproduction was substantial. In the previous study using similar methods, the common slope was  $-6.810 \pm 1.718$  and the 95% confidence intervals overlapped with those of the common slope of  $-5.341$  in this study. Using the three control costs and market values described by Gordy et al. (2019), a range of economic injury levels was calculated for the common slope of  $-5.34$  of the combined yield-maximum aphid density regressions of susceptible hybrids. For these hybrids, economic injury levels varied from 27 aphids per leaf when the grain market value was high and the cost of control was low, to 71 aphids per leaf when the grain value was low and cost of control was high. This range is in agreement with the results presented by Gordy et al. (2019).

For partially resistant hybrids, 42 of the 49 hybrids across all location-years had a non-significant (flat) slope ( $P > 0.05$ ), indicating no yield loss as aphid density increased (Table 4-4). The highest mean aphid densities observed across the 42 hybrids ranged from 6 to 352 aphids per leaf (Table 4-4). For five of the 49 instances, significant regressions appeared to be coincidental and unrelated to aphid density. For example, BH 4100 (Gainesville TX 2016 and Rosenberg TX 2016) showed an upward yield response to increased aphid density. In these cases, growing conditions and soil moisture were very good. We note that six location-years (CC16, GV16, RB16, CC17, GV17, RB18B) did not have a known susceptible hybrid that demonstrated significant yield loss resulting from sugarcane aphid damage. However, for those susceptible hybrid by

location-year combinations (n=11), the range of observed maximum aphid density was 67 to 1,025 aphids per leaf, with a mean of 454 aphids per leaf. For the partially resistant hybrids across the same location-years (n=24), the mean maximum aphid density was 104 aphids per leaf. Across all location-year combinations, the reduction of maximum aphid density on resistant hybrids ranged from 25-99%, compared to a susceptible hybrid at the same location, with a mean maximum aphid density reduction of 81% (Table 4-4). In addition to the overall reduction of maximum aphid density on partially resistant hybrids, doubling time for sugarcane aphid populations on partially resistant hybrids was up to 6.4-fold higher than doubling time on known susceptible hybrids. 14 of 18 hybrids evaluated as partially resistant demonstrated yield stability in location-years where a companion susceptible hybrid experienced significant yield decline. The remaining four of 18 hybrids evaluated as partially resistant demonstrated a large reduction in maximum aphid density or an increase in aphid population doubling time, when compared to the susceptible companion hybrid at the same location, supporting their designation as partially resistant to sugarcane aphid. When considering yield stability, reduction in maximum aphid density, or increased aphid population doubling time, 47 of the 49 partially resistant hybrid by location-year combinations demonstrated suppression of natural aphid populations under full-season field conditions. Overall, for all partially resistant hybrid location-years, 47 of 49 showed no yield loss resulting from increased density of sugarcane aphid. These results support that the purported hybrids with partial sugarcane aphid resistance available from several seed companies have yield stability in multiple field settings across a range of sugarcane aphid densities.



**Table 4-4 Yield-aphid density estimate linear regressions for maximum aphid density (MAD) from regional field experiments on partially resistant and susceptible sorghum hybrids in the southern U.S., 2016-2018**

Year	Location	Hybrid, S/R	n	Intercept <sup>a</sup>	Slope <sup>a</sup>	R <sup>2</sup>	P value	Mean MAD <sup>c</sup>	Max. MAD <sup>d</sup>	% Red. Mean MAD <sup>e</sup>	% Red. Max. MAD <sup>f</sup>
2016	Corpus Christi TX	SP7715,R	16	1724 [740.8,2707]	-38.77 [-122.9,43.32]	0.0653	0.3396	8	39	93	89
		BH4100,R	16	1922 [1244,2600]	66.02 [-5.368,137.4]	0.2194	0.0673	5	35	95	90
		DK3888,S	16	294.1 [-205.5,793.7]	3.454 [-1.349,8.257]	0.1452	0.1453	80	258	--	--
		DK3707,R	16	461.6 [85.75,834.5]	5.752 [-3.442,14.95]	0.1139	0.2010	29	115	72	68
		DK4807,R	16	641.1 [174.5,1108]	-14.28 [-62.53,33.97]	0.0280	0.5359	6	25	94	93
2016	Winnsboro LA	DK5367,S	16	1755 [1012,2498]	-4.512 [-10.10,1.075]	0.1765	0.1052	103	363	--	--
		DK3707,R	16	5305 [4918,5691]	-0.173 [-9.150,8.803]	0.0001	0.9676	35	75	76	78
		DK3888,S	16	5515 [5007,6023]	-2.714 [-5.920,0.439]	0.1962	0.0857	138	264	--	--
2016	Rosenberg TX	REV9782,S	16	5901 [5537,6264]	-4.670 [-6.687,2.652]	0.6378	0.0002	144	344	--	--
		BH4100,R	16	2653 [1975,3332]	4.846 [0.333,9.359]	0.2748	0.0371	134	293	45	39
		DK3707,R	16	2595 [1999,3191]	3.814 [-0.238,7.867]	0.2255	0.0631	128	283	48	41
		DK3888,S	16	3235 [2690,3779]	-0.376 [-2.334,1.583]	0.0119	0.6871	244	483	--	--
		DK4807,R	16	3520 [2874,4165]	2.171 [-7.919,12.26]	0.0150	0.6515	54	147	78	70
2016	Gainesville TX	DK5367,S	16	3670 [3100,4239]	-1.173 [-3.323,0.976]	0.0892	0.2612	237	397	--	--
		SP7715,R	16	3034 [2370,3699]	2.886 [-1.326,7.098]	0.1336	0.1638	140	278	43	42
		DK3707,R	16	2427 [886.1,3967]	2.083 [-13.72,17.88]	0.0057	0.7815	83	175	76	78
		DK3888,S	16	1595 [753.9,2437]	0.842 [-1.982,3.668]	0.0284	0.5326	244	648	--	--
		DK4807,R	16	2955 [1683,4227]	1.525 [-13.86,16.91]	0.0032	0.8346	64	218	82	73
		DK5367,S	16	1981 [828.8,3133]	0.108 [-2.771,2.988]	0.0005	0.9369	348	810	--	--
		SP7715,R	16	2237 [688.5,3785]	9.019 [-18.70,36.74]	0.0336	0.4967	49	92	86	89
2016	Monticello AR	BH4100,R	16	1875 [598.5,3151]	37.74 [0.550,74.93]	0.2528	0.0471	29	62	92	92
		DK3707,R	16	4555 [3735,5375]	1.844 [-11.42,15.11]	0.0063	0.7700	59	105	81	86
		DK3888,S	16	4037 [2664,5410]	-4.108 [-8.110,-0.107]	0.2572	0.0449	311	727	--	--
		83P17,R	16	5184 [4552,5817]	-5.418 [-12.61,1.777]	0.1571	0.1286	82	138	74	81
2016	Florence SC	84P80,S	16	5043 [7279,5087]	-6.000 [-9.149,-2.851]	0.5440	0.0011	201	533	--	--
		DK3707,R	16	2748 [2199,3297]	-2.295 [-10.66,6.075]	0.0241	0.5659	62	132	64	70
		DK3888,S	16	2440 [1754,3126]	-3.705 [-6.349,1.061]	0.3923	0.0094	174	434	--	--
2017	Corpus Christi TX	BH4100,R	8	2954 [2626,3281]	-16.14 [-82.68,50.39]	0.0555	0.5744	4	9	97	97
		DK5367,S	8	865.1 [-219,1949]	2.470 [-4.820,9.760]	0.1028	0.4388	118	288	--	--
		83P56,R	8	2639 [1310,3968]	-26.17 [-72.02,19.69]	0.2453	0.2121	22	70	81	76
		TX430,S	7	2158 [993.9,3323]	-6.264 [-15.82,3.288]	0.3624	0.1526	105	207	--	--
		TX2783,R	6	960.9 [115.0,1807]	-3.658 [-28.46,21.15]	0.0402	0.7032	24	77	80	73
		SP73B12,R	8	848.0 [43.30,1653]	63.84 [-84.11,211.8]	0.1567	0.3317	5	11	96	96
		SP7715,R	6	553.1 [45.91,1060]	-3.453 [-17.99,11.08]	0.0981	0.5456	20	83	83	71
		W7051,R	8	653.6 [351.3,955.8]	32.60 [-3.065,68.26]	0.4547	0.0667	6	22	95	93
		W844E,R	6	2095 [847.3,3342]	-2.548 [-38.08,32.98]	0.0098	0.8519	25	78	78	73
		WE44,R	8	3073 [1334,4811]	-97.18 [-259.2,64.83]	0.2642	0.1925	10	19	92	93

**Table 4, continued**

Year	Location	Hybrid, S/R	n	Intercept <sup>a</sup>	Slope <sup>a</sup>	R <sup>2</sup>	P value	Mean MAD <sup>c</sup>	Max. MAD <sup>d</sup>	% Red. Mean MAD <sup>e</sup>	% Red. Max. MAD <sup>f</sup>		
2017	Gainesville TX	OL242,R	12	6454 [4485,8422]	-107.8 [-212.4,-3.336]	0.3458	0.0443	17	37	90	91		
		DK3707,R	12	4445 [3203,5687]	-1.518 [-42.62,39.59]	0.0007	0.9360	26	56	85	87		
		BH3822,S	12	4654 [3136,6173]	-5.208 [-12.71,2.288]	0.1933	0.1527	173	421	--	--		
		BH4100,R	12	5983 [3832,8135]	-133.3 [-382.4,115.9]	0.1244	0.2609	8	13	95	97		
		W625Y,R	12	5995 [4633,7356]	-35.26 [-61.83,-8.687]	0.4664	0.0144	44	98	75	77		
		SP73B12,R	12	3595 [583.7,6606]	2.519 [-267.7,272.7]	0.000	0.9838	10	19	94	96		
		83P56,R	12	4035 [3308,4763]	-9.053 [-14.18,-3.924]	0.6074	0.0028	111	316	36	25		
2017	Rosenberg TX	TX2783,R	8	4865 [4303,5426]	-5.819 [-16.06,4.418]	0.2438	0.2137	40	102	74	78		
		BH4100,R	8	5273 [4327,5917]	-26.50 [-68.33,15.33]	0.2860	0.1721	12	30	92	93		
		TX430,S	8	4967 [4437,5497]	-2.220 [-15.03,10.59]	0.0291	0.6864	33	67	--	--		
		DK4807,R	8	4804 [3819,5789]	1.089 [-35.89,38.07]	0.0009	0.9449	20	51	87	89		
		DK5367,S	8	4642 [3910,5374]	-4.793 [-7.829,-1.758]	0.7133	0.0083	156	454	--	--		
		W7051,R	8	4176 [3788,4564]	-2.569 [-25.27,20.13]	0.0126	0.7911	11	41	93	91		
		SP7715,R	8	5818 [4629,7007]	-109.6 [-248.2,28.89]	0.3846	0.1009	7	15	95	97		
		83P56,R	8	5072 [4442,5701]	1.113 [-10.74,12.97]	0.0087	0.8259	30	145	81	68		
		W844E,R	8	5471 [5010,5933]	-20.27 [-54.28,13.75]	0.2616	0.1951	12	20	92	96		
		2018	Rosenberg TX A	DK4513,S	8	3461 [2724,4200]	-4.008 [-6.177,-1.838]	0.7730	0.0040	252	591	--	--
DK4707,R	8			3099 [2632,3567]	-1.612 [-13.18,9.959]	0.0190	0.7448	29	83	90	89		
481,R	8			3806 [3397,3820]	1.870 [-9.616,13.36]	0.0258	0.7041	15	37	95	95		
DK5367,S	8			3456 [2550,4362]	-3.701 [-5.898,-1.503]	0.7389	0.0062	303	730	--	--		
SP78M30,R	8			2973 [2452,3494]	-3.275 [-16.33,9.774]	0.0591	0.5617	31	83	90	89		
83P56,R	8			3358 [3754,3962]	-5.504 [-10.62,0.388]	0.5360	0.0389	85	226	72	69		
3247,R	8			3158 [2801,3515]	-2.170 [-17.00,12.66]	0.0209	0.7325	17	49	94	93		
74GB17,R	8			3456 [3133,3779]	-7.873 [-17.71,1.958]	0.3902	0.0978	27	63	91	91		
2018	Rosenberg TX B			SP78M30,R	12	4288 [3840,4737]	-12.81 [-23.33,2.298]	0.4243	0.0217	22	100	87	90
				DK5367,S	12	2005 [1180,2831]	-0.062 [-2.472,2.348]	0.0003	0.9552	173	1025	--	--
		DK4707,R	12	3593 [2754,4433]	13.06 [-21.88,47.99]	0.0648	0.4244	16	56	91	95		
		74GB17,R	12	4206 [3651,4761]	-2.486 [-6.347,1.375]	0.1707	0.1819	74	352	57	66		
		DK4513,S	12	3003 [1917,4089]	0.609 [-3.205,40413]	0.0125	0.7295	166	676	--	--		
		3247,R	12	3794 [3090,4497]	7.776 [-23.34,38.90]	0.0301	0.5899	11	71	94	93		
		83P56,R	11	2631 [768.8,4493]	4.852 [-36.14,45.85]	0.0079	0.7950	25	83	86	92		
		481,R	11	2833 [970.9,4696]	391.8 [-155.5,879.1]	0.2176	0.1481	3	6	98	99		
All Susceptible <sup>b</sup>		--	88	4429 [3999,4860]	-5.341 [-6.848,-3.833]	0.3658	<0.0001	--	--	--	--		

<sup>a</sup> Parameter estimate followed by 95% confidence interval.

<sup>b</sup> All Susceptible indicates the common point estimates from the regression analysis for the combined regression of all susceptible hybrids with significant ( $p < 0.05$ ) individual hybrid-location year regressions. For MAD there were seven hybrids across five location years (n=88).

<sup>c</sup> Mean maximum aphid density across all experimental plots for the hybrid at a given location-year.

<sup>d</sup> Highest maximum aphid density observed in any plot for the hybrid at a given location-year.

#### **4.4. Discussion**

Based on data using 25 sorghum hybrids (18 resistant, 7 susceptible) across 11 location-years, field-based population growth rates of sugarcane aphid were calculated for partially resistant and susceptible sorghum hybrids. The range of economic injury levels and the average economic threshold of 40 aphids per leaf based on maximum aphid density during infestation of vegetative growth proposed by Gordy et al. (2019) was reaffirmed for susceptible hybrids added to the data set from this experiment.

For commercial hybrids reported to be partially sugarcane aphid resistant, yield stability was maintained across peak aphid densities that ranged from 6 to 352 aphids per leaf. Wilson et al. (2020) found lower sugarcane aphid densities on resistant grain sorghum hybrids compared to susceptible hybrids, and Lahiri et al. (2020) showed that yield of sugarcane aphid resistant grain sorghum hybrids remained stable compared to susceptible hybrids. Although these partially resistant hybrids do not allow for calculation of an economic injury level based on yield-aphid density regressions (Pedigo et al. 1986), it is clear that many of these hybrids maintain good yield at aphid densities much higher than the range of economic injury levels of 27 to 71 aphids per leaf for susceptible hybrids (Gordy et al. 2019). The lack of yield response to aphid density may indicate some of these hybrid's ability to tolerate or compensate for injury due to aphid feeding across a wide range of environmental conditions when aphid densities were relatively high. In contrast, low aphid population growth and aphid densities may reflect aphid suppression by some of these hybrids. The variation in aphid densities with no yield loss observed suggests that one or more resistance mechanism (i.e. tolerance,

antibiosis, antixenosis) may be present in the partially resistant hybrids (Stout 2013, Sharma 1993). Paudyal et al. (2020) reported that selected resistant sorghums tolerate some physiological effects of sugarcane aphid feeding when artificially infesting plants. Categorizing these commercial resistant hybrids into groupings of resistance mechanisms or identifying specific resistant traits would require pedigree information of the hybrids and additional breeding and genetic study (Stout 2013).

Given the range of environmental conditions and locations represented, these results suggest that growers have a high likelihood of finding a partially resistant hybrid that is locally adapted to specific sorghum-producing areas of the U.S. represented in our study as well as other areas given the wide geographic range of the locations used here (Table 4-1). Sugarcane aphid populations increase considerably above the currently used economic threshold of 40 aphids per leaf for susceptible hybrids (Gordy et al. 2019) in some of these partially resistant commercial hybrids. Therefore, there is value to have background information on  $r$ ,  $DT$ , and  $\lambda$  of the sorghum hybrid planted when monitoring sugarcane aphid. If aphids are observed when growing these partially aphid resistant hybrids, consideration of supplemental insecticide use may be warranted when aphid densities exceed the average population sizes experienced for each of these hybrids (Table 4-4). If the hybrid and its background to sustain sugarcane aphid populations are not known, it would be prudent to monitor for any unusual leaf decay associated with aphid densities once aphid densities exceed the range of 27 to 71 aphids per leaf used as the economic injury level for susceptible hybrids (Gordy et al. 2019).

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## 5. EVALUATION OF INSECTICIDES FOR EFFICACY AND SPRAY TIP AND APPLICATION VOLUME COMBINATIONS FOR SPRAY COVERAGE IN MANAGEMENT OF SUGARCANE APHID (HEMIPTERA: APHIDIDAE) IN GRAIN SORGHUM

### 5.1. Introduction

One recent management issue for grain sorghum, *Sorghum bicolor* L., is the sugarcane aphid, *Melanaphis sacchari* Zehntner. Since first detection of outbreaks on sorghum in 2013, this pest has contributed to yield loss and harvest issues in sorghum producing areas in Texas and across sorghum producing regions of the U.S. and Mexico (Bowling et al. 2016). Because of the sugarcane aphid's potential for population increase and potential to damage aphid-susceptible sorghum, insecticides have been used to manage the aphid beginning in 2014 (Bowling et al. 2016). Expected proportion of control is critical to evaluating insecticide effectiveness and is one of the factors that is used in calculating an economic threshold to guide insecticide use.

The active ingredient and spray coverage of a foliar-applied insecticide affect expected proportion of insect control (Zehnder and Speese 1991, Farias et al. 2020). Active ingredients can vary in effectiveness to reduce a population of insects. A wide variety of spray tips including flat fan, even flat fan, extended range (XR), drift guard, air induction (AI), dual fan, and others are currently available for grower use. Historically, insect control has employed hollow cone nozzles to maximize spray coverage by directing spray in all directions and achieving small droplet sizes to reach deep into the crop canopy (Welty et al. 1995). It has also been shown that increased

spray volume and pressure result in increased coverage and canopy penetration, however, this is often not practiced as increased spray volume decreases production efficiency (Legleiter and Johnson 2016, Welty et al. 1995). With the adoption of some herbicides that require larger droplet sizes (in-crop uses of dicamba and 2,4 D) and spray volumes (glufosinate), some growers will often tank-mix and co-apply insecticides, or will use these coarse spray tips for all applications to reduce time delays in changing spray nozzles.

To provide guidance on insecticide selection for *M. sacchari* management, labeled and unlabeled insecticides were evaluated for reduction of *M. sacchari* populations in commercially grown grain sorghum. For the purpose of demonstration, coverage (i.e., canopy penetration in commercial grain sorghum) was measured using several combinations of spray tips and spray volumes.

## **5.2. Methods and Materials**

### **5.2.1. Insecticide Efficacy**

Field experiments were conducted near Rosenberg, Texas from 2015 through 2017, and at the Texas A&M AgriLife Research and Extension Center, Corpus Christi, Texas in 2017. All trials were conducted on DeKalb DKS 53-67 grain sorghum, a known aphid-susceptible hybrid (Brewer et al. 2017) The experiment conducted near Rosenberg in 2015 was performed on ratoon sorghum, while all other experiments were conducted on first-crop sorghum. *Melanaphis sacchari* populations were monitored in order to implement trials when populations were near economic thresholds (Gordy et al. 2019).

The experiments conducted in Rosenberg in 2015 and 2016 were replicated three times. The experiments conducted in Rosenberg and Corpus Christi in 2017 were replicated four times. The experiment conducted in Rosenberg in 2017 was arranged in a completely randomized design while all other experiments were arranged in a randomized complete block. Experiments consisted of plots with lengths ranging from 7.62 to 12.19 m by four rows spaced 1.02 m apart. Spray tips, spray volume, and insecticides varied by experiment (Tables 5-1 and 5-2). All applications were made using a CO<sub>2</sub>-powered backpack sprayer with four tips spaced 51 cm apart. Insecticide applications were applied when *M. sacchari* densities were detected at or above economic thresholds (Gordy et al. 2019). Agronomic management practices for all trials adhered to standard regional practices (e.g., Trostle and Fromme 2010).

Aphid measurements were conducted prior to insecticide application and every three to seven days following application. In each plot, the lower-most green leaf and the upper-most unfurled leaf below the flag leaf were examined on 10 randomly selected plants, for a total of 20 leaves per plot. Aphids were counted up to 50 per leaf, after which the populations were estimated by 10s up to 250, 50s up to 500, and 100s thereafter. The total number of aphids per plot was divided by 20 to calculate the average aphid density per plot. Insect count data were transformed ( $\log(n+1)$ ) to meet assumptions of normality. Analysis of variance was performed using PROC MIXED in SAS 9.3 (SAS Institute, Cary, NC) and means were separated using Tukey-Kramer HSD for all pairwise comparisons.

**Table 5-1 Experimental design and application details for insecticide efficacy studies conducted near Rosenberg and Corpus Christi, Texas, 2015-2017**

<b>Location, Year</b>	<b>Plot Design</b>	<b>Replications</b>	<b>Treatments</b>	<b>Spray Date</b>	<b>Spray Volume</b>	<b>Spray Tip</b>	<b>Experiment Code</b>
Rosenberg, 2015	RCB	3	5	09/04/2015	126 L/ha	TeeJet TTJ60-11002	A
Rosenberg, 2016	RCB	3	14	09/07/2016	131 L/ha	TeeJet AI110015	B
Rosenberg, 2017	CRD	4	5	06/01/2017	131 L/ha	TeeJet AI110015	C
Corpus Christi, 2017	RCB	4	7	11/21/2017	126 L/ha	TeeJetTTJ60-11002	D

**Table 5-2 Active ingredient, IRAC group, formulation, and manufacturer of insecticides evaluated on *Melanaphis sacchari* on grain sorghum in Texas, 2015-2017**

Trade Name	Active Ingredient(s)	IRAC Group <sup>a</sup>	Formulation <sup>b</sup>	Manufacturer	Experiment <sup>c</sup>
Baythroid XL	Beta-cyfluthrin	3A	L	Bayer; St. Louis, MO	A,C
Carbine 50WG	Flonicamid	9C	WG	FMC; Philadelphia, PA	B
Couraze	Imidicloprid	4A	F	Chemnova; Research Triangle Park, NC	B
Dimethoate 4EC	Dimethoate	1B	EC	Helena; Collierville, TN	B
Endigo ZC	Lambda-cyhalothrin, Thiamethoxam	3A, 4A	ZC	Syngenta; Greensboro, NC	A,B,C,D
Fulfill	Pymetrozine	9B	WDG	Adama; Raleigh, NC	B
Lorsban 4E	Chlorpyrifos	1B	E	Corteva; Indianapolis, IN	B
PFR-97WG	<i>Isaria fumosorosea</i> Apopka Strain 97	n/a	WDG	Certis; Columbia, MD	B
Sefina	Afidopyropen	9D	DC	BASF; Research Triangle Park, NC	B,D
Sivanto Prime	Flupyradifurone	4D	L	Bayer; St. Louis, MO	A,B,C,D
Transform WG	Sulfoxaflor	4C	WG	Corteva; Indianapolis, IN	A,B,C,D

<sup>a</sup> As defined by the Insecticide Resistance Action Committee mode of action classification.

<sup>b</sup> Formulation based on product label.

<sup>c</sup> Experiment code as listed in Table 5-1.

### 5.2.2. Spray Tip and Spray Volume

Experiments were performed in a commercial grain sorghum field using grower equipment. Nine spray tips or tip configurations (Table 5-3) were evaluated. They were fitted on a spray boom covering three rows. Six spray tips were spaced 51 cm apart across three 1.02 m rows. Final spray volumes evaluated were 65 and 112 L/ha (7 and 12 gallons per acre, respectively). A John Deere 4720 self-propelled sprayer applied the 65 and 112 L/ha spray volumes using pressures of 207 and 276 KPa at speeds of 25.1 and 16.6 km/h, respectively. The applications were made on May 16, 2017. The air temperature was 28-29°C, humidity was 50%, and the wind speed was 8-13 km/h, direction was perpendicular to the direction of rows of the boot-stage sorghum. F

Plots consisted of two rows of sorghum 75 m long, with one row between plots. Plots were placed in the middle a sorghum field with 1.02 m row spacing. Measurements at four canopy positions were used to evaluate spray coverage. Canopy positions were the base of the plant stalk (1) (approximately 10 cm above ground), the base of the lower-most green leaf (2) (approximately 25 cm above ground), the base of the second leaf below the upper-most unfolded leaf (3) (approximately 50 cm above ground), and on the apex of the upper-most unfolded leaf (4) (approximately 80 cm above ground) (Fig. 5-1). Water sensitive cards (Syngenta, Basel, Switzerland) were placed at each of the designated plant canopy position of five plants per plot (interior of two treated rows) within the spray path of the tractor (Fig. 5-2).

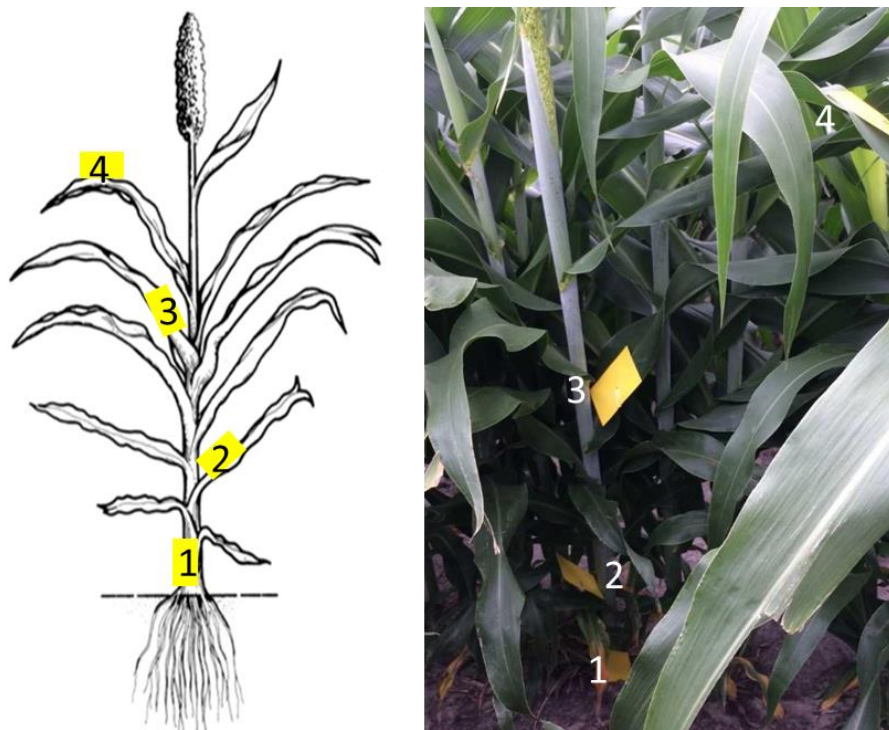
**Table 5-3 Spray tip, droplet size classification, orientation, and Manufacturer of spray tips evaluated for percent coverage and canopy penetration in grain sorghum in Texas, 2016-2017**

<b>Brand</b>	<b>Spray Tip</b>	<b>Droplet Size at 206-276 KPa<sup>a</sup></b>	<b>Orientation<sup>b</sup></b>	<b>Manufacturer</b>
Agrotop	TC 110-04	Coarse	Standard	Agrotop; Obertraubling, Germany
Greenleaf	DF 04	Coarse	Standard	Greenleaf; Covington, LA
Greenleaf	DF 04	Coarse	Alternating	Greenleaf; Covington, LA
Greenleaf	TADF 04	Very Coarse - Coarse	Standard	Greenleaf; Covington, LA
Greenleaf	TADF 04	Very Coarse - Coarse	Alternating	Greenleaf; Covington, LA
TeeJet	AIXR 110-04	Extra Coarse	Standard	Teejet; Glendale Heights, IL
TeeJet	AI 3070-04	Extra Course – Very Coarse	Standard	Teejet; Glendale Heights, IL
TeeJet-ConeJet	TXR 8004VK	Fine	Standard	Teejet; Glendale Heights, IL
TeeJet-TurboTeeJet	TTJ60-11004	Coarse	Standard	Teejet; Glendale Heights, IL

<sup>a</sup> Droplet size based on data provided by manufacturer, equivalent to 30-40 pounds per square inch.

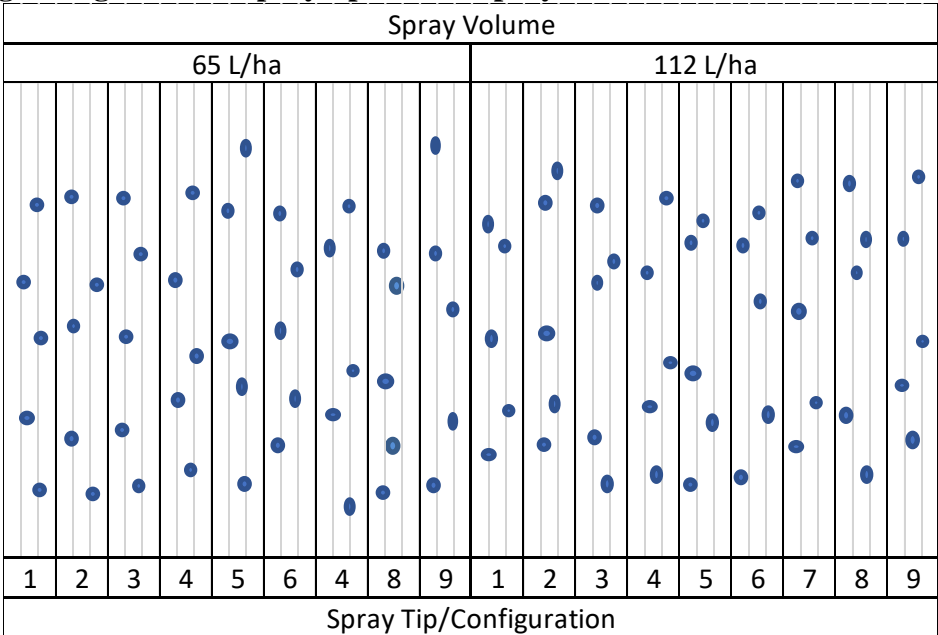
<sup>b</sup> Standard indicates all spray tips pointing the same direction; alternating indicates tips alternating spray direction (forward/backward) to increase angles of spray.

**Figure 5-1 Placement of water sensitive cards at 1) base of plant stalk (10 cm), 2) base of the lowest green leaf (25 cm), 3) base of the second leaf below the upper-most unfolded leaf (50 cm), and 4) on the apex of the upper-most unfolded leaf (80 cm) to determine canopy penetration of spray droplets**





**Figure 5-2 Generalized layout water sensitive cards within plots to determine spray coverage using different spray tips and of spray volumes**



\*Faint gray lines within spray tip/configuration indicate rows, dots indicate example of distribution of five plants used for placement of water sensitive cards.

Following spray application, water sensitive cards were allowed to dry for one hour before being collected. Each card was processed using a Penpower WorldCard color business card scanner and the image was assessed using DepositScan software (USDA-ARS, Wooster, OH). These data were for demonstration purposes and to evaluate if an experiment with replication of the treatments was warranted. Only descriptive statistics were calculated and provided.

### **5.3. Results and Discussion**

#### **5.3.1. Insecticide Efficacy**

Field evaluations demonstrated acceptable efficacy of several products to reduce populations of sugarcane aphids to levels below the economic threshold of 40 aphids per leaf (Gordy et al. 2019). For the experiment conducted near Rosenberg in 2015, significant treatment differences were observed at three (d.f.= 4, 9;  $F=22.7$ ;  $P<0.001$ ) seven (d.f.= 4, 9;  $F=84.98$ ;  $P<0.001$ ), and 14 (d.f.= 4, 9;  $F=15.92$ ;  $P<0.001$ ) days after application (Table 5-4). There were no differences among treatments prior to application (d.f.=4,9;  $F=2.16$ ;  $P=0.155$ ) or at 19 (d.f.= 4, 9;  $F=1.27$ ;  $P=0.35$ ) or 23 (d.f.= 4, 9;  $F=3.36$ ;  $P=0.06$ ) days after application. The experiment conducted near Rosenberg in 2016 showed significant differences in aphid populations among treatments at four (d.f.= 13,27;  $F=8.42$ ;  $P<0.001$ ) and nine (d.f.= 13,27;  $F=4.81$ ;  $P<0.001$ ) days after application. There were no differences in aphid populations prior to application (d.f.= 13,27;  $F=1.52$ ;  $P=0.1732$ ) or at 16 (d.f.= 13,27;  $F=0.99$ ;  $P=0.50$ ) days after application (Table 5-5). The experiment near Rosenberg in 2017 had significant treatment effects at all post-

application assessment timings (d.f.= 4,15;  $F>5.94$ ;  $P<0.01$ ) (Table 5-6). The trial performed near Corpus Christi in 2017 also had significant treatment effects at all post-application assessment timings (d.f.= 6, 20;  $F>4.88$ ;  $P <0.01$ ) (Table 5-7).

In all trials, at least one treatment was different from the untreated check ( $p<0.05$ ).

The two commercial standards, Transform WG and Sivanto Prime, demonstrated population reduction of >93% at six or seven days after application (compared to the untreated check), in all four location years. Baythroid XL, a pyrethroid, did not reduce populations of *M. sacchari* in either of the experiments in which it was included (Rosenberg 2015 and 2017). Endigo ZC, a premix of thiamethoxam and lambda-cyhalothrin (a neonicotinoid and pyrethroid, respectively), provided similar efficacy to that of Transform WG and Sivanto Prime, with >89% population reduction at six or seven days after application. Evaluated in a single location-year, Carbine 50WG (0.3 kg/ha), Lorsban 4E (1.17 L/ha) + Dimethoate 1.17 (L/ha), PFR-97 WG (1.12 kg/ha) + Transform WG (0.07 kg/ha), and Couraze (0.62 L/ha) showed reduction of *M. sacchari* populations, compared to the untreated check. Treatments that did not provide reduction of aphid populations included Carbine 50WG 0.2 kg/ha, Carbine 50WG (0.2 kg/ha) + Dimethoate (1.17 L/ha), PFR-97 WG (1.12 kg/ha), and Fulfill (0.365 L/ha) + Kinetic (0.29 L/ha). For all except for the Rosenberg 2017 experiment, Transform WG, Sivanto Prime, and Endigo ZC provided comparable reduction of *M. sacchari* populations at two, three, or four days after application. For Rosenberg in 2017, only the Transform treatment reduced the *M. sacchari* population below that of the untreated check at four days after application. In the two trials tested (Rosenberg 2016 and Corpus Christi

**Table 5-4 Mean aphid per leaf as an indicator of efficacy of selected insecticides applied to commercial grain sorghum near Rosenberg, Texas in 2015**

Product, Rate	Days After Application										
	0 <sup>a</sup>	3 <sup>ab</sup>		7 <sup>ab</sup>		14 <sup>ab</sup>		19 <sup>a</sup>	23 <sup>a</sup>	Yield <sup>bc</sup>	
Transform WG, 0.07 kg/ha	160.7	76.4	B	23.2	B	343.3	AB	340.1	493.8	15.0	B
Sivanto Prime, 0.29 L/ha	145.3	55.5	B	4.3	C	114.9	C	86	101.5	154.0	A
Endigo ZC, 0.365 L/ha	85.5	73.3	B	11.3	BC	257.3	BC	169.6	327.8	89.3	AB
Baythroid XL, 0.175 L/ha	135.5	219	A	364.1	A	852.2	A	117.7	65.8	17.0	B
Untreated Check	131.3	300.8	A	370.7	A	689.5	A	120.7	72.3	25.3	B

<sup>a</sup> Data were transformed ( $\log(n+1)$ ) to meet assumptions of normality; non-transformed means shown

<sup>b</sup> Means followed by the same letter are not significantly different ( $\alpha=0.05$ ) as determined by Tukey-Kramer HSD for all pairwise comparisons, conducted only when  $F$ -test was significant.

<sup>c</sup> Yield measured in number of heads per plot.

**Table 5-5 Mean aphid per leaf as an indicator of efficacy of selected insecticides applied to commercial grain sorghum near Rosenberg, Texas in 2016**

Treatment	Days After Application					16 <sup>a</sup>	Yield <sup>c</sup>
	0 <sup>a</sup>	4 <sup>ab</sup>			9 <sup>ab</sup>		
Carbine 50WG 0.2 kg/ha	114.4	23.9	ABC	8.4	BC	0.0	1291
Carbine 50WG 0.2 kg/ha + Dimethoate 1.17 L/ha	55.1	18.3	ABC	9.2	ABC	0.4	1289
Lorsban 4E 1.17 L/ha + Dimethoate 1.17 L/ha	74.9	10.4	BCD	6.7	BC	0.0	2386
Carbine 50WG 0.3 kg/ha	85.5	3.6	BCD	1.9	BC	0.2	2578
Sivanto 0.29 L/ha	85.5	2	CD	1.2	BC	0.0	2536
Transform WG 0.07 kg/ha	54.4	0.5	D	2.4	BC	0.0	2242
PFR-97 WG 1.12 kg/ha	71.8	47.3	AB	6.6	AB	0.1	1414
PFR-97 WG 1.12 kg/ha + Transform WG 0.07 kg/ha	49.6	4.8	BCD	4.3	BC	0.0	2467
Endigo ZC 0.365 L/ha	49.5	1.1	D	3.8	BC	0.0	2445
Couraze 0.62 L/ha	71.1	9.3	BCD	4.6	BC	0.1	2305
Fulfill 0.365 L/ha + Kinetic 0.29 L/ha	41.6	84.4	ABC	66	ABC	0.0	2392
Sefina 0.20 L/ha + MSO 0.5% v/v	105.3	32.8	ABC	1.7	BC	0.0	2332
Sefina 0.40 L/ha + MSO 0.5% v/v	61.1	8.7	BCD	0.9	C	0.0	2470
Untreated Check	72.7	142.6	A	133.8	A	2.5	1078

<sup>a</sup> Data were transformed (log(n+1)) to meet assumptions of normality; non-transformed means shown.

<sup>b</sup> Means followed by the same letter are not significantly different ( $\alpha=0.05$ ) as determined by Tukey-Kramer HSD for all pairwise comparisons, conducted only when *F*-test was significant.

<sup>c</sup> Yield measured in kilograms per hectare.

2017), Sefina appeared to require longer to reduce populations to levels below the untreated check (Tables 5-5 and 5-7). Additionally, in the Corpus Christi 2017 trial, Sefina treatments did not reduce populations to levels equivalent to those achieved by Transform WG or Sivanto Prime (Table 5-7).

Duration of population reduction by selected products varied by location-year. For example, in Rosenberg in 2015, *M. sacchari* populations showed a resurgence to levels above the economic threshold (mean of 114 aphids per leaf for lowest treatment) by 14 days after application for all products. Similarly, in Corpus Christi (2017), aphid populations began to rebound by 15 days after application for treatments of Transform WG, while populations remained low for other products. For the other two location-years, Rosenberg 2016 and 2017, populations for all plots, including the untreated check fell to very low levels by 16 and 24 days after application, respectively.

Beneficial insect counts, including predators and parasitoids of *M. sacchari*, were not reported here. Compatibility with beneficial insect populations, including pollinators and predators of other sorghum pests, should be considered when selecting an insecticide or other method of control for *M. sacchari* in grain sorghum. Some insecticides have label restrictions in place, specifically for pollinators (e.g. Transform WG and Sivanto).

### **5.3.2. Spray Tip and Spray Volume**

For canopy position, the upper canopy (position 4, 80 cm above the ground) received the highest spray coverage: an average of 8.51% of the area of the water sensitive cards.

Spray coverage for all other canopy levels were similar with the middle canopy (position

3, 50 cm above the ground) receiving 1.45% coverage, the lower canopy (position 2, 25 cm above the ground) receiving 1.20% spray coverage, and the plant base (position 1, 10 cm above the ground) receiving 1.23% spray coverage. Although crop architecture is quite different, these findings are similar to observations in soybean by Farias et al. (2020), where higher coverage was found in the upper canopy compared to the middle and lower canopy. Mean spray coverage across all canopy levels and spray volumes ranged from 2.58% for the hollow cone spray tip to 4.18% for the air induction extended range (AIXR) spray tip.

Previous research demonstrated that tips producing fine droplets produced 9.5 times the driftable fine droplets (<100 microns) compared to a similar air induction tip producing coarse to extra coarse droplets. Specifically, 3.8% of the fine spray vs. 0.396% of the coarse spray when applying at a pressure of 207 Kpa (McGinty et al. 2016), the same pressure used for the 65 L/ha application volume in this study. Considering the crosswind experienced during spray application in this experiment, tips providing larger spray droplets may deliver more spray lower into the canopy as the small droplets can be blown horizontally off-target and intercepted by the upper canopy.

When considering spray volume, mean spray coverage for 65 and 112 l/ha was 2.75% and 3.56% across all canopy positions, respectively. For canopy positions two and three, mean spray coverage was 0.86% and 1.79% for 65 and 112 l/ha, respectively. Similar results were observed by Sharpe et al. (2017) where doubling spray volume from 187 to 375 L/ha increased spray coverage in the lower canopy of strawberry by 81%.

**Table 5-6 Mean aphid per leaf as an indicator of efficacy of selected insecticides applied to grain sorghum near Rosenberg, Texas in 2017**

Treatment	Days After Application													
	0 <sup>a</sup>	4 <sup>ab</sup>		7 <sup>ab</sup>		10 <sup>ab</sup>		14 <sup>ab</sup>		19 <sup>ab</sup>		24 <sup>ab</sup>	Yield <sup>c</sup>	
Transform WG 0.07 kg/ha	67.5	0.6	C	0.8	C	2.9	B	7.6	B	1.3	B	0.3	B	4819
Sivanto Prime 0.29 L/ha	48.2	19.3	ABC	0.1	C	0.8	B	0.4	B	0.1	B	0.1	B	5059
Endigo ZC 0.365 L/ha	73	5.3	BC	16.1	B	0.9	B	2.4	B	1	B	0.2	B	4761
Baythroid XL 0.175 L/ha	61.1	66.9	A	139.8	A	261.6	A	143.7	A	150.8	A	15.8	A	3812
Untreated Check	54.2	35.3	AB	145.6	A	288.6	A	194.8	A	128.5	A	15.5	AB	3544

<sup>a</sup> Data were transformed (log(n+1)) to meet assumptions of normality; non-transformed means shown.

<sup>b</sup> Means followed by the same letter are not significantly different ( $\alpha=0.05$ ) as determined by Tukey-Kramer HSD for all pairwise comparisons, conducted only when *F*-test was significant.

<sup>c</sup> Yield measured in kilograms per hectare.



**Table 5-7 Mean aphid per leaf as an indicator of efficacy of selected insecticides applied to grain sorghum near Corpus Christi, Texas in 2017**

Treatment	Days After Application			
	0 <sup>a</sup>	2 <sup>ab</sup>	6 <sup>ab</sup>	15 <sup>ab</sup>
Sefina 0.33 L/ha	351.5	190.3 AB	49.5 BC	11.8 B
Sefina 0.44 L/ha	345.0	375.8 A	79.3 B	64.0 B
Endigo ZC 0.365 L/ha	365.5	155.3 ABC	16.3 CD	9.5 B
Transform WG 0.07 kg/ha	309.3	36.5 C	2.5 EF	33.8 AB
Sivanto Prime 0.29 L/ha	268.5	107.5 BC	11.3 DE	15.5 B
Transform WG 0.105 kg/ha	311.5	60.5 C	1.75 F	27.3 B
Untreated Check	262.3	467.5 A	580.8 A	680.3 A

<sup>a</sup> Data were transformed ( $\log(n+1)$ ) to meet assumptions of normality; non-transformed means shown.

<sup>b</sup> Means followed by the same letter are not significantly different ( $\alpha=0.05$ ) as determined by Tukey-Kramer HSD for all pairwise comparisons, conducted only when *F*-test was significant.

When considering relative concentration of the insecticide in 65 and 112 L/ha spray volumes, the latter would deliver 18% more active ingredient to the lower and middle canopy of the sorghum crop as was evaluated here. Zehdner and Speese (1991) demonstrated that increasing spray volume from 140 to 560 L/ha resulted in increased control of Colorado potato beetle in potato.

#### **5.4. Conclusions**

Management of *M. sacchari* immediately after its introduction relied on labeled insecticide products such as Lorsban 4E, which offered low to moderate control with a pre-harvest interval of 30-60 days, depending on the rate used (Buntin and Roberts 2016, Black et al. 2018, Buntin et al. 2018). Within two years, flupyradifurone and sulfoxaflor received labels for use against the aphid and these two products have been shown to be efficacious against the aphid (Buntin et al. 2018). Additional products and active ingredients evaluated here, including Sefina and the thiamethoxam component of Endigo ZC, have demonstrated acceptable control. The solo formulation of thiamethoxam is under the label name Centric and has shown good control in several other evaluations of *M. sacchari* (Black et al. 2018, Buntin et al. 2018), but is not currently labeled for use in sorghum.

There was no observed difference in the effect of spray tip on spray coverage. Spray tips that deliver fine droplets (<225 microns, e.g. hollow cone) and coarse to extra coarse droplets (>325 microns, e.g. air induction, dual fan, etc.) were shown to provide the same coverage and thus should provide comparable efficacy with a given insecticide.

Increasing spray volume from 65 to 112 L/ha resulted in increased spray coverage in grain sorghum.

The data presented here suggest increased spray volumes may improve canopy penetration and spray coverage in grain sorghum. Additional testing of spray volumes, including higher volumes than those tested here, and efficacy tests specifically comparing spray tips and volumes should be conducted to confirm that comparable spray coverage between fine and course droplet producing spray tips translates to comparable control by insecticides. The results from the efficacy trials presented here and performed by others (Buntin and Roberts 2016, Buntin et al. 2018, Black et al. 2018) confirm several labeled insecticides that provide good control of *M. sacchari* in grain sorghum. Insecticides should only be applied based on economic thresholds. Other management considerations including use of resistant hybrids, conservation of natural enemies, and other cultural practices should be considered (Michels and Burd 2007) and mitigated with strategically applied insecticides when economic thresholds are exceeded.

## 5.5. References

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## 6. SUMMARY

The intent of the research described here was to estimate the aphid density-yield loss relationship, develop economic thresholds appropriate to a range of hybrids and environmental conditions, and evaluate a proportion-tally threshold approach as an alternate to a density-based threshold. Additional objectives were to evaluate partially aphid-resistant hybrids for aphid growth potential and yield stability across a range of conditions and demonstrate efficacy of insecticides, and evaluate application technologies for selective use as guided by economic thresholds.

### **6.1. Development of economic thresholds in susceptible grain sorghum**

This research included a wide range of geography, environmental conditions, production practices, cropping seasons, and sugarcane aphid population ranges. Economic thresholds ranged from 19 to 132 aphids per leaf, with mean economic injury levels of 37 aphids per leaf for environments where aphid populations increase relatively rapidly and 102 aphids per leaf for environments where populations increase relatively slowly. Some susceptible hybrids appear to avoid yield loss when aphids exceed the suggested range of ETs if there is sufficient soil moisture and aphid populations build up quickly and rapidly decline. Modifications of these thresholds are appropriate based on changes in commodity price, management costs, and desired outcomes of their respective sorghum pest management program. However, without site-specific knowledge of what regulates slow- or fast-growing aphid populations and given cost and market price variability of the system, a 40 aphid per leaf threshold is most prudent to

use across the range of hybrid, environmental, and market conditions experienced in this study.

## **6.2. Binomial-based tally thresholds as an alternative to density-based thresholds**

The use of an infestation proportion based on a tally threshold appears to be a suitable alternative to the density-based approach of economic thresholds. Of the three tally threshold levels evaluated here, utilizing a tally threshold proportion of 0.16 for a tally threshold of >50 aphids per leaf provided the best match of decisions when compared to the established density-based threshold of 40 aphids per leaf. For differences in aphid reproductive potential, market prices, and control costs as noted in chapter 2, the regression parameters estimated may be used by a pest manager to calculate the tally threshold proportion for any applicable density-based economic threshold. The tally threshold proportion approach also required half the time to complete a 100-leaf sample, compared to estimating aphid densities of a 100 leaf sample. This work supports use of a tally threshold as a time-saving alternative to the density-based threshold for use in hybrids that are aphid susceptible, and those with varying levels of resistance.

## **6.3. Field assessment of aphid doubling time and yield of sorghum susceptible and partially resistant to sugarcane aphid**

Based on data using 25 sorghum hybrids (18 resistant, 7 susceptible) across 11 location-years, field-based population doubling time for sugarcane aphid populations on

partially resistant hybrids was greatly increased, ranging from 1.3- to 6.4-fold, compared to doubling time on known susceptible hybrids. Economic injury levels and economic thresholds based on maximum aphid density during infestation of vegetative growth were reaffirmed for susceptible hybrids. For resistant hybrids, yield was variable but stable across a range of conditions and economic injury level could not be estimated. Across the 18 partially resistant hybrids evaluated in this study, considerable variation in sugarcane aphid density and aphid population doubling time were observed. The yield stability of the 47 of 49 partially resistant hybrids across a range of aphid densities and environmental conditions suggests that one or more resistance mechanism (i.e. tolerance, antibiosis, antixenosis) may be present in the partially resistant hybrids evaluated here.

#### **6.4. Evaluation of insecticides for efficacy and spray tip and application volume for spray coverage in management of sugarcane**

Data indicate there are several labeled and unlabeled insecticides that effectively reduce populations of *M. sacchari* by >90%. Sivanto Prime, Transform WG, and Sefina are labeled and efficacious against *M. sacchari*. Sivanto Prime and Transform WG generally reduce populations more quickly than Sefina, and Sivanto Prime typically provides the greatest duration of control. There was no observed difference in the effect of spray tip on spray coverage. Spray tips that deliver fine droplets and coarse to extra coarse droplets were shown to provide the same coverage and thus should provide comparable efficacy with a given insecticide with systemic activity. When considering



only the middle and lower canopy of sorghum, canopy penetration as assessed by spray coverage increases when spray volume is increased from 65 to 114 L/ha.

## **6.5. Other Considerations**

The work summarized here provides valuable tools for management of *M. sacchari* in grain sorghum. The yield-aphid density relationship between the aphid and sorghum was estimated across a wide range of geographies, growing conditions, hybrids, and aphid pressure. Economic injury levels and economic thresholds were calculated considering a range of control costs and market values and proportion tally thresholds were evaluated as an alternative to the density-based threshold. Partially resistant grain sorghum hybrids demonstrated yield stability across a wide range of conditions and aphid densities. This work provides pest managers the ability to calculate an economic injury level, economic threshold, and tally-threshold proportion that fits with their production practices and goals. It also provides support for use of partially resistant hybrids showing yield stability across a wide range of environments and conditions. Finally, it demonstrates efficacy of insecticides for selective use as determined by economic thresholds.

To provide support for the regional application of these results, a robust set of data was collected and curated through collaboration with other university researchers throughout the southern U.S. In doing so, economic injury level could be calculated and a best estimate of a density-based economic threshold for sugarcane aphid on susceptible sorghum hybrids relevant across south, central, and north central Texas, and additional

southern states where data was congregated (Gordy et al. 2019). With a similar degree of collaboration, evaluation of partially resistant commercial sorghum hybrids confirmed yield stability across a wide range of aphid densities and growing conditions. Furthermore, a tally-based threshold was demonstrated to be a suitable substitute for the density-based approach while providing time savings.

With full-time employment as a County Extension Agent – Agriculture and Natural Resources within Texas A&M AgriLife Extension at the beginning of this endeavor, an additional goal was to provide information to growers, consultants, and other stakeholders throughout the process. This was achieved by providing updates at grower field days and regional industry meetings, as well as providing relevant information in newsletters and in annual summaries to growers (in the form of result demonstration reports) and stakeholders (in the form of annual research summaries).

Throughout most of the experiments conducted in Texas, incidence of predators and parasitoids of *M. sacchari* were also recorded. Future work could consider how field surroundings influence aphid infestation, population growth, and aphid population modulation. The role of the beneficial insect complex on regulation of sugarcane aphid populations and assessment of the extent to which predators and parasitoids adjust to *M. sacchari* populations is currently being investigated, led by members and collaborators of the field crops entomology laboratory of Texas A&M AgriLife Research, Corpus Christi, Ashleigh Faris and Blake Elkins.

Early work on *M. sacchari* showed a correlation between population growth and environmental conditions. Observations of excellent growing conditions coincided with

reduced yield decline, even when aphids were at levels that caused yield decline under different conditions. Additional investigation into the role of abiotic factors in population regulation – specifically, precipitation and temperature, and sorghum management factors including irrigation and fertility, could help to better understand what contributes to higher or lower aphid population growth rates and other factors that contribute to reduced yield loss.

Since taking on this project, the annual incidence and relative impact of sugarcane aphid has seemed to decline, with fewer fields requiring treatment and the duration of infestations declining. Although anecdotal, I hypothesize that this is the result of a combination of events. First, I believe the adoption of partially resistant hybrids across much of South Texas and along the Gulf Coast has resulted in an overall reduction in total aphids that are able to migrate northward throughout the growing season. Second, I think that the beneficial insect complex (i.e. predators such as syrphids, chrysomelids, and chrysopids and parasitoids) has adjusted to some degree, to respond more quickly to aphid colonization, once it occurs. I believe these two parts (resistant hybrids and beneficial insects) work together, the former enabling the latter to have a greater impact. Third, throughout my research, the propensity for populations to rapidly decline has seemed to increase. Based on other aphid pests of row crops, specifically cotton aphid in cotton, entomopathogenic fungi with the ability to cause an epizootic event could have become more prevalent in these areas. This combination of resistant hybrid adoption by producers, seasonal adjustment by beneficial insects, and the potential for epizootic events may have led to an overall decrease in economic

damage by the sugarcane aphid over the last couple years. However, additional research is needed to confirm or negate these hypothesized changes in frequency and intensity of aphid infestation and damage since its introduction.

APPENDIX

**Table A1 Infestation proportion – mean density linear regression parameters for tally thresholds of >25, >50, and >100 aphids per leaf, for individual field experiments on grain sorghum in Texas, 2015-2018**

Year	Location	Tally	Slope <sup>a,b</sup>	Intercept <sup>a</sup>	R <sup>2</sup>	P value
2015	Corpus Christi, TX	>25	0.0023 [0.0021, 0.0025]	0.0697 [0.0470, 0.0923]	0.7642	<0.0001
		>50	0.0020 [0.0019, 0.0022]	0.0086 [0.0095, 0.0435]	0.8191	<0.0001
		>100	0.0015 [0.0015, 0.0016]	0.0027 [-0.0072, 0.0125]	0.8855	<0.0001
2015	Rosenberg, TX	>25	0.0013 [0.0011, 0.0015]	0.3205 [0.2713, 0.3697]	0.5308	<0.0001
		>50	0.0014 [0.0012, 0.0016]	0.2164 [0.1754, 0.2574]	0.8003	<0.0001
		>100	0.0015 [0.0014, 0.0016]	0.0633 [0.0371, 0.0895]	0.8292	<0.0001
2016	Corpus Christi, TX	>25	0.0032 [0.0031, 0.0034]	0.0083 [0.0048, 0.0118]	0.7087	<0.0001
		>50	0.0023 [0.0022, 0.0024]	0.0018 [-0.0005, 0.0042]	0.7349	<0.0001
		>100	0.0016 [0.0015, 0.0016]	-0.0001 [-0.0015, 0.0012]	0.7989	<0.0001
2016	Rosenberg, TX	>25	0.0038 [0.0035, 0.0040]	0.1344 [0.1142, 0.1565]	0.5847	<0.0001
		>50	0.0034 [0.0033, 0.0035]	0.0357 [0.0247, 0.0467]	0.8942	<0.0001
		>100	0.0026 [0.0025, 0.0026]	-0.0123 [-0.0179, -0.0067]	0.9110	<0.0001
2016	Gainesville, TX	>25	0.0028 [0.0027, 0.0030]	0.0783 [0.0661, 0.0904]	0.5779	<0.0001
		>50	0.0025 [0.0024, 0.0026]	0.0228 [0.0163, 0.0294]	0.7836	<0.0001
		>100	0.0020 [0.0020, 0.0021]	-0.0002 [-0.0034, 0.0029]	0.9129	<0.0001
2017	Corpus Christi, TX	>25	0.0031 [0.0029, 0.0032]	0.0065 [0.0036, 0.0094]	0.7547	<0.0001
		>50	0.0023 [0.0022, 0.0024]	0.0002 [-0.0019, 0.0023]	0.7672	<0.0001
		>100	0.0018 [0.0017, 0.0018]	-0.0022 [-0.0035, -0.0009]	0.8361	<0.0001
2017	Rosenberg, TX	>25	0.0032 [0.0031, 0.0034]	0.0207 [0.00153, 0.0262]	0.6994	<0.0001
		>50	0.0028 [0.0027, 0.0029]	0.0046 [0.0017, 0.0075]	0.8534	<0.0001
		>100	0.0021 [0.0020, 0.0021]	-0.0013 [-0.0030, 0.0003]	0.9115	<0.0001
2017	Gainesville, TX	>25	0.0037 [0.0035, 0.0040]	0.0479 [0.0357, 0.0600]	0.7039	<0.0001
		>50	0.0033 [0.0031, 0.0034]	0.0146 [0.0062, 0.0229]	0.7941	<0.0001
		>100	0.0024 [0.0023, 0.0025]	-0.0054 [-0.0097, -0.0010]	0.8849	<0.0001
2018	Rosenberg, TX(A)	>25	0.0023 [0.0021, 0.0024]	0.0454 [0.0340, 0.01567]	0.6092	<0.0001
		>50	0.0021 [0.0020, 0.0022]	0.0175 [0.00967, 0.0254]	0.7343	<0.0001
		>100	0.0018 [0.0018, 0.0019]	0.0015 [-0.0035, 0.0066]	0.8370	<0.0001
2018	Rosenberg, TX(B)	>25	0.0018 [0.0017, 0.0020]	0.0394 [0.0225, 0.0563]	0.6014	<0.0001
		>50	0.0018 [0.0016, 0.0019]	0.0237 [0.0114, 0.0360]	0.7242	<0.0001
		>100	0.0016 [0.0016, 0.0017]	0.0106 [0.0020, 0.0192]	0.8228	<0.0001
2018	Gainesville, TX	>25	0.0051 [0.0046, 0.0055]	0.0075 [0.0014, 0.0137]	0.7360	<0.0001
		>50	0.0039 [0.0036, 0.0041]	-0.0043 [-0.0081, -0.0006]	0.8113	<0.0001
		>100	0.0021 [0.0018, 0.0024]	-0.0031 [-0.0074, 0.0012]	0.4979	<0.0001

<sup>a</sup> Parameter estimate followed by 95% confidence interval.

<sup>b</sup> The slope reflects the incremental increase of the proportion of leaves with >X aphids (25, 50, or 100) for each aphid present on a leaf.

**Table A2 Infestation proportion – mean density polynomial regression parameters for tally thresholds of >25, >50, and >100 aphids per leaf, for individual field experiments on grain sorghum in Texas, 2015-2018**

Year	Location	Tally	Slope $a^a$		Slope $b^{a,b}$		Intercept <sup>a</sup>		$R^2$	$P$ value
2015	Corpus Christi, TX	>25	-5.7e <sup>-6</sup>	±5.1e <sup>-7</sup>	0.0043	±1.9e <sup>-4</sup>	0.0259	±0.0097	0.8606	<0.0001
		>50	-4.1e <sup>-6</sup>	±3.9e <sup>-7</sup>	0.0035	±1.5e <sup>-4</sup>	-0.0051	±0.0074	0.8880	<0.0001
		>100	-2.2e <sup>-6</sup>	±2.4e <sup>-7</sup>	0.0023	±9.0e <sup>-5</sup>	-0.0141	±0.0045	0.9222	<0.0001
2015	Rosenberg, TX	>25	-3.4e <sup>-6</sup>	±3.2e <sup>-7</sup>	0.0035	±2.1e <sup>-4</sup>	0.1643	±0.0242	0.7225	<0.0001
		>50	-2.9e <sup>-6</sup>	±2.6e <sup>-7</sup>	0.0032	±1.8e <sup>-4</sup>	0.0834	±0.0199	0.7943	<0.0001
		>100	-1.7e <sup>-6</sup>	±1.7e <sup>-7</sup>	0.0026	±1.2e <sup>-4</sup>	-0.0161	±0.0133	0.8927	<0.0001
2016	Corpus Christi, TX	>25	-1.1e <sup>-5</sup>	±5.0e <sup>-7</sup>	0.0052	±1.1e <sup>-4</sup>	0.0011	±0.0015	0.8020	<0.0001
		>50	-6.3e <sup>-6</sup>	±3.5e <sup>-7</sup>	0.0035	±7.5e <sup>-5</sup>	0.0024	±0.0011	0.8021	<0.0001
		>100	-2.7e <sup>-6</sup>	±2.2e <sup>-7</sup>	0.0021	±4.6e <sup>-5</sup>	-0.0019	±0.0006	0.8267	<0.0001
2016	Rosenberg, TX	>25	-2.1e <sup>-5</sup>	±7.4e <sup>-7</sup>	0.0099	±2.3e <sup>-4</sup>	0.0459	±0.0079	0.8251	<0.0001
		>50	-1.1e <sup>-5</sup>	±3.3e <sup>-7</sup>	0.0067	±1.0e <sup>-4</sup>	-0.1201	±0.0035	0.9417	<0.0001
		>100	-2.3e <sup>-6</sup>	±2.8e <sup>-7</sup>	0.0033	±8.7e <sup>-5</sup>	-0.0220	±0.0030	0.9204	<0.0001
2016	Gainesville, TX	>25	-8.4e <sup>-6</sup>	±2.5e <sup>-7</sup>	0.0066	±1.2e <sup>-4</sup>	0.0352	±0.0044	0.8082	<0.0001
		>50	-5.1e <sup>-6</sup>	±1.1e <sup>-7</sup>	0.0048	±5.6e <sup>-5</sup>	0.0032	±0.0020	0.9312	<0.0001
		>100	-2.2e <sup>-6</sup>	±7.0e <sup>-8</sup>	0.0030	±3.2e <sup>-5</sup>	-0.0112	±0.0011	0.9593	<0.0001
2017	Corpus Christi, TX	>25	-3.7e <sup>-6</sup>	±7.7e <sup>-7</sup>	0.0038	±1.6e <sup>-4</sup>	0.0045	±0.0015	0.7625	<0.0001
		>50	2.3e <sup>-6</sup>	±5.6e <sup>-7</sup>	0.0019	±1.1e <sup>-4</sup>	0.0014	±0.0011	0.7724	<0.0001
		>100	-3.9e <sup>-6</sup>	±3.2e <sup>-7</sup>	0.0011	±6.4e <sup>-5</sup>	0.0000	±0.0006	0.8648	<0.0001
2017	Rosenberg, TX	>25	-1.3e <sup>-5</sup>	±4.6e <sup>-7</sup>	0.0065	±1.6e <sup>-4</sup>	0.0076	±0.0021	0.8324	<0.0001
		>50	-5.9e <sup>-6</sup>	±2.4e <sup>-7</sup>	0.0047	±8.3e <sup>-5</sup>	-0.0030	±0.0011	0.9262	<0.0001
		>100	-2.6e <sup>-6</sup>	±1.6e <sup>-7</sup>	0.0029	±5.4e <sup>-5</sup>	-0.0046	±0.0007	0.9387	<0.0001
2017	Gainesville, TX	>25	-1.2e <sup>-5</sup>	±7.2e <sup>-7</sup>	0.0068	±2.0e <sup>-4</sup>	0.0141	±0.0051	0.8298	<0.0001
		>50	-8.1e <sup>-6</sup>	±5.1e <sup>-7</sup>	0.0053	±1.4e <sup>-4</sup>	0.0078	±0.0036	0.8747	<0.0001
		>100	-2.6e <sup>-6</sup>	±3.1e <sup>-7</sup>	0.0030	±8.9e <sup>-5</sup>	-0.0124	±0.0022	0.9016	<0.0001
2018	Rosenberg, TX(A)	>25	-8.0e <sup>-6</sup>	±2.5e <sup>-7</sup>	0.0065	±1.4e <sup>-4</sup>	0.0136	±0.0035	0.8685	<0.0001
		>50	-5.6e <sup>-6</sup>	±1.8e <sup>-7</sup>	0.0051	±9.8e <sup>-5</sup>	0.0048	±0.0024	0.9121	<0.0001
		>100	-3.2e <sup>-6</sup>	±1.3e <sup>-7</sup>	0.0035	±7.4e <sup>-5</sup>	-0.0110	±0.0018	0.9234	<0.0001
2018	Rosenberg, TX(B)	>25	-3.7e <sup>-6</sup>	±2.0e <sup>-7</sup>	0.0044	±1.5e <sup>-4</sup>	0.0179	±0.0059	0.8211	<0.0001
		>50	-3.0e <sup>-6</sup>	±1.1e <sup>-7</sup>	0.0039	±8.7e <sup>-5</sup>	0.0059	±0.0034	0.9204	<0.0001
		>100	-2.3e <sup>-6</sup>	±6.0e <sup>-8</sup>	0.0032	±4.9e <sup>-5</sup>	-0.0027	±0.0019	0.9671	<0.0001
2018	Gainesville, TX	>25	-4.5e <sup>-5</sup>	±7.1e <sup>-6</sup>	0.0078	±4.8e <sup>-4</sup>	-0.0017	±0.0032	0.7824	<0.0001
		>50	-7.4e <sup>-7</sup>	±4.8e <sup>-6</sup>	0.0039	±3.3e <sup>-4</sup>	0.0045	±0.0022	0.8114	<0.0001
		>100	-2.5e <sup>-6</sup>	±5.2e <sup>-6</sup>	0.0006	±3.5e <sup>-4</sup>	0.0020	±0.0023	0.5519	<0.0001

<sup>a</sup> Parameter estimate from polynomial regression where  $y=ax^2+bx$ +intercept, followed by the Standard Error.

<sup>b</sup> The slope reflects the incremental increase of the proportion of leaves with >X aphids (25, 50, or 100) for each aphid present on a leaf.